Synthesis of carba analogues of intermediates in the shikimate pathway

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SYNTHESIS OF CARBA ANALOGUES OF INTERMEDIATES IN THE SHIKIMATE PATHWAY

Submitted by

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for the degree of Ph.D.

of the University of Bath

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Dedicated to my Mother and Father

for their love and support.

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ABSTRACT

The shikimate pathway is the major biosynthetic route by which plants and micro-organisms produce the aromatic amino acids and a plethora of other natural products. The details of this pathway are discussed in Chapter 1, together with a review of the recent syntheses of shikimic acid and of later intermediates in the shikimate pathway.

Our synthesis of analogues of shikimic acid, which are of interest as potential enzyme inhibitors, is described in Chapter 2. Starting from the Diels-Alder adducts of *N*-alkyloxycarbonyl-1,2-dihydropyridines and methyl acrylate, these were ring opened upon treatment with base to afford 5-(*N*-alkyloxycarbonylamino-methyl)cyclohexa-1,3-diene-1-carboxylates. Deprotection yielded the amino acid (±)-5-homogabaculine, the 5-homologue of naturally occurring gabaculine.

The *cis*-hydroxylation of these dienes, either through osmylation or through the use of a 'wet' Prévost reaction, gave 5-homoshikimate derivatives. Deamination was achieved *via* displacement of a disulphonimide to afford (±)-5-homoshikimic acid. An improved route to this compound involved the conversion of a trifluoroacetamide derivative to the corresponding acetate *via* nitrosation. These studies also led to a range of 5-substituted shikimic acid analogues.

The synthesis of a carba analogue of 5-enolpyruvylshikimic acid, was accomplished by addition of an α-acrylate synthon to a 5-iodomethyl shikimate.

Finally, the syntheses of homochiral products were studied utilising a chiral acrylate in the initial Diels-Alder cycloaddition. The use of Lewis acid catalysts induced decomposition of the dihydropyridine used, however, the thermal reaction afforded the diastereomeric cycloaddition adducts which were separable. Thus, both enantiomers of a key intermediate in the earlier syntheses became available.

Full experimental details for the preparation of these compounds are given in Chapter 3.

ABBREVIATIONS

Ac - acetyl

AIBN - azobis(isobutyronitrile)

ATP - adenosine triphosphate

aq. - aqueous

Bn - benzyl

BOC - *tert*-butoxycarbonyl

BSA - bis(trimethylsilyl)acetamide

Bu - butyl

t-Bu - *tert*-butyl

Bz - benzoyl

C.I. - chemical ionisation

m-CPBA - *m*-chloroperbenzoic acid

15-crown-5 - 1,4,7,10,13-pentaoxacyclopentadecane

18-crown-6 - 1,4,7,10,13,16-hexaoxacyclooctadecane

DABCO - 1,4-diazabicyclo[2.2.2]octane

DAHP - 3-deoxy-D-arabinoheptulosonic acid 7-phosphate

DAST - diethylaminosulphur trifluoride

DBN - 1,5-diazabicyclo[4.3.0]non-5-ene

DBU - 1,8-diazabicyclo[5.4.0]undec-7-ene

DCC - dicyclohexylcarbodiimide

2D COSY - two dimensional correlated spectroscopy

d.e. - diastereomeric excess

DEAD - diethyl azodicarboxylate

DHP - 1,2-dihydropyridine

DMA - 3,5-dimethoxyaniline

DMAP - 4-dimethylaminopyridine

DMF - *N*,*N*-dimethylformamide

DMPU - 1,3-dimethyl-3,4,5,6-tetrahydro-2(1*H*)-pyrimidinone

DMSO - dimethyl sulphoxide

E.I. - electron ionisation

EPSP - 5-enolpyruvylshikimate-3-phosphate

Et - ethyl

FAB - fast atom bombardment

GABA - γ-aminobutyric acid

h - hour

HMPA - hexamethylphosphoramide

HOMO - highest occupied molecular orbital

IR - infrared

J - coupling constant

LDA - lithium diisopropylamide

LUMO - lowest unoccupied molecular orbital

Me - methyl

MEM - 2-methoxyethoxymethyl

min - minute

morph-DAST - morpholinosulphur trifluoride

m.p. - melting point

NAD^+, NADH - nicotinamide adenine dinucleotide, reduced form

$NADP^+$, NADPH - nicotinamide adenine dinucleotide phosphate, reduced form

NBS - *N*-bromosuccinimide

NMO - *N*-methylmorpholine *N*-oxide

NMR - nuclear magnetic resonance

n.O.e - nuclear Overhansen effect

Ns - *p*-nitrobenzenesulphonyl

Nu - nucleophile

PCC - pyridinium chlorochromate

PEP - phosphoenolpyruvate

Ph - phenyl

PPTS - pyridinium *p*-toluenesulphonate

Pr - propyl

i-Pr - isopropyl

Py - pyridine

R_F - retention factor

S-3-P - shikimate-3-phosphate

S_N1 - substitution nucleophilic unimolecular

S_N2 - substitution nucleophilic bimolecular

TBDMS - *tert*-butyldimethylsilyl

Tf - trifluoromethylsulphonyl (triflyl)

TFA - trifluoroacetic acid

TFAA - trifluoroacetic anhydride

THF - tetrahydrofuran

TLC - thin layer chromatography

TMS - Trimethylsilyl

TPAP - tetra *n*-propylammonium perruthenate

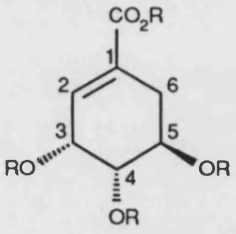
Ts - *p*-toluenesulphonyl (tosyl)

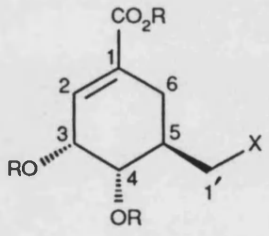
p-TSA - *p*-toluenesulphonic acid

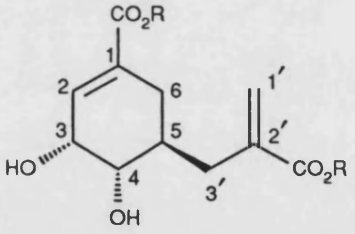
NOMENCLATURE

The nomenclature of cyclohexene and cyclohexane compounds referred to in this thesis, is based on shikimic acid nomenclature, even though this may not necessarily conform to IUPAC convention. This permits analysis of any compound without reference to the nomenclature for that particular compound, and furthermore, allows direct comparison of NMR data.

The numbering system employed labels the carboxylate substituted carbon as C-1 and proceeds anticlockwise around the ring, through the double bond. More highly substituted derivatives are named as depicted below. All other compounds are named in accordance with IUPAC rules.







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CHAPTER ONE

INTRODUCTION

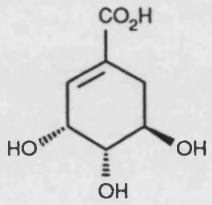
CHAPTER ONE

INTRODUCTION

1.1 The Shikimate Pathway

1.1.1 Introduction

The glucose-derived shikimate pathway and the acetate-derived polyketide pathway are the major routes for the biosynthesis of aromatic compounds in plants and micro-organisms.[1] The former leads to the aromatic amino acids, L-phenylalanine, L-tyrosine and L-tryptophan and is named after a key intermediate in the pathway, shikimic acid (**1**).



(**1**)

Shikimic acid was first isolated by Eykmann,[2] in 1885, from the fruit of *Illicium religiosum* and it is from the Japanese name for this plant, *shikimi-no-ki*, that the name for this compound was derived. The structure and absolute stereochemistry was later proven by the work of Fischer and Dangschat,[3] Freudenberg[4] and Karrer.[5]

The true importance of shikimic acid was not appreciated until the 1950's with the work of Davis.[6] Mutant strains of *Escherichia coli* and *Aerobacter aerogenes* were produced, which had a growth requirement for five aromatic compounds (L-phenylalanine, L-tyrosine, L-tryptophan, *p*-aminobenzoic acid and *p*-hydroxybenzoic acid). Certain mutants were found to accumulate shikimic acid,

while other mutants, blocked at a different point in the pathway, were found to be able to utilise shikimic acid to replace the aromatic substrates. These observations identified shikimic acid as a common precursor for each of these aromatic compounds. Further studies by Davis, Sprinson,[7] Gibson[8] and subsequent workers, revealed a well defined series of eight intermediates, leading from glucose, first to shikimic and then to chorismic acid.

1.1.2 The Common Pathway

The main stem of the shikimate pathway from carbohydrate to chorismate is generally referred to as the common pathway (**Figure 1.1**). The mechanisms for each stage remain debateable, however, the sequence of intermediates is clear.

Oxidation of glucose by the pentose phosphate pathway affords D-erythrose-4-phosphate (**3**) and, by glycolysis, phosphoenolpyruvate (PEP, **2**). The condensation of these produces the seven-carbon saccharide 3-deoxy-D-arabino-heptulosonic acid (DAHP, **4**). The mechanism of the DAHP synthase reaction has been the subject of some speculation.[1d] Originally a concerted process was postulated (**Figure 1.2**), in which nucleophilic attack at the phosphorus atom of PEP results in cleavage of the P-O bond and generation of a reactive enol pyruvate anion which can rapidly add to (**3**).

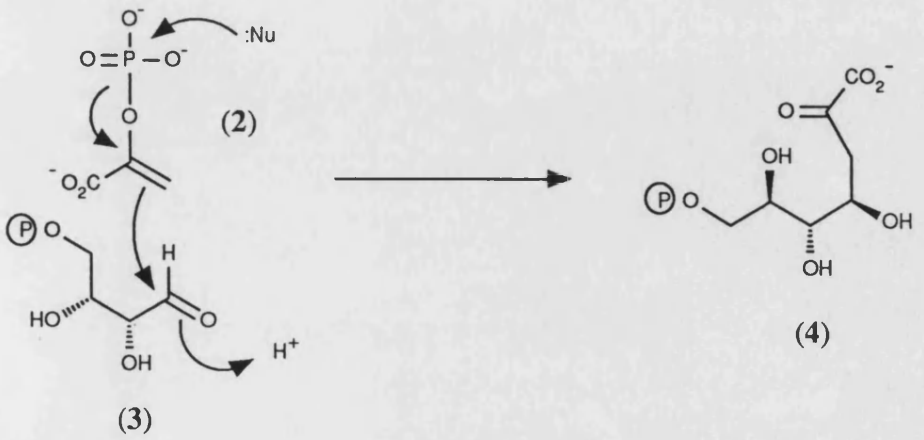


Figure 1.2 Postulated Mechanism for DAHP Synthase

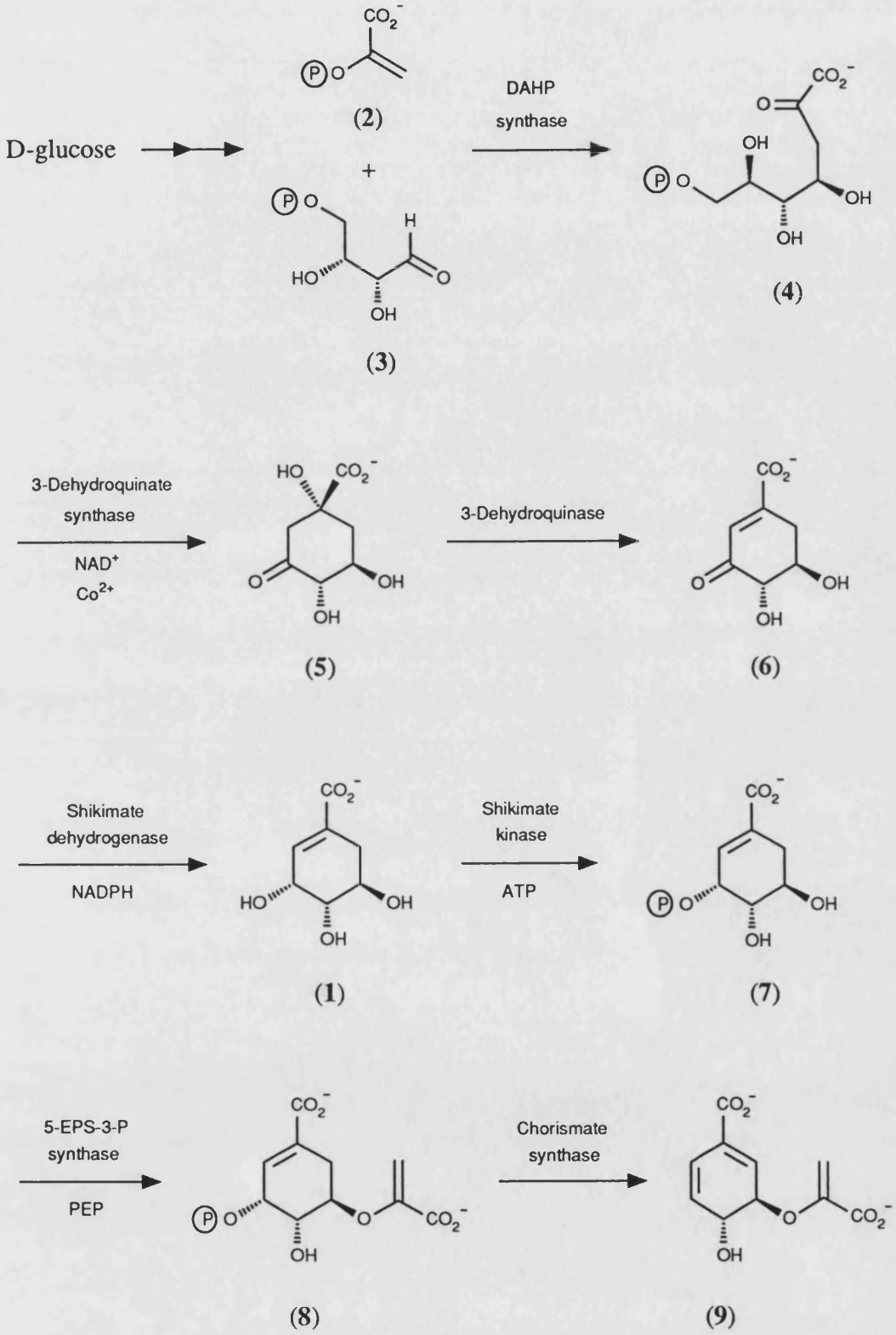


Figure 1.1 The Common Pathway.

However, later kinetic studies suggested that DAHP synthase operated by a 'ping-pong' mechanism, in which one of the products of the reaction was released before both substrates had bound to the enzyme. Since PEP tended to stabilize the enzyme against denaturation, it seemed logical to suggest that an enzyme bound enolpyruvyl complex was formed initially and phosphate released. Experiments with ^{18}O labelled substrates showed that the reaction involved C-O as opposed to P-O bond cleavage. On the basis of these findings an alternative mechanism was proposed (**Figure 1.3**), in which the substrate PEP was first transferred to a nucleophilic group on the enzyme, such as a carboxyl group. Elimination of phosphate leaves an enolpyruvyl-enzyme complex, which undergoes acyl-oxygen cleavage and initiates the aldol condensation with the second substrate (**3**).

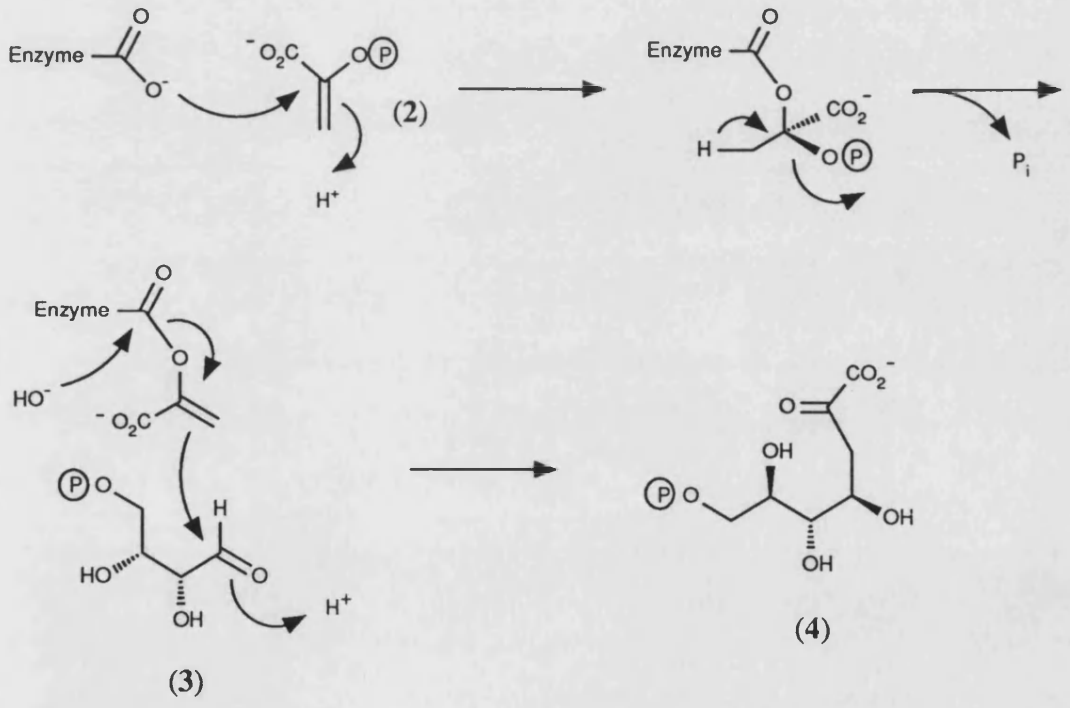
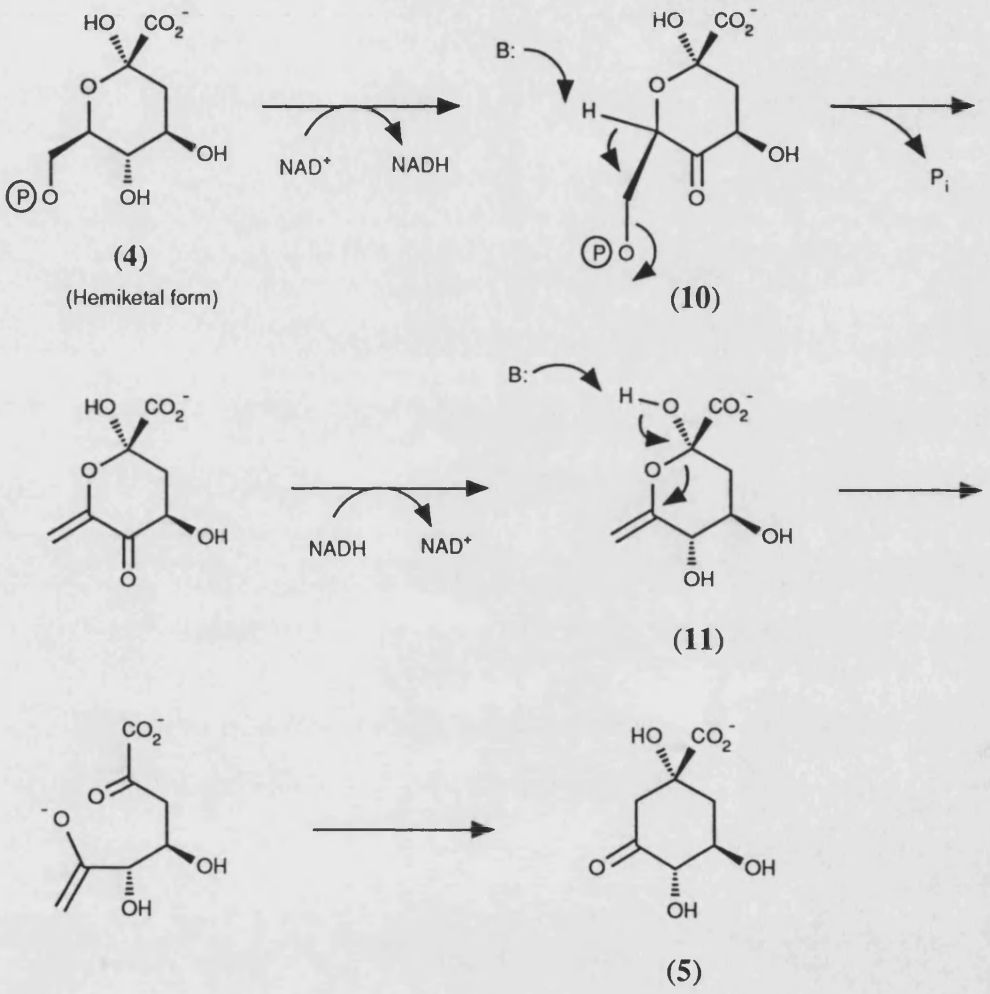


Figure 1.3 Postulated 'Ping-pong' Mechanism for DAHP Synthase.

3-Dehydroquinate synthase catalyses the conversion of DAHP (**4**) to 3-dehydroquinate (**5**) and requires NAD^+ and a divalent metal cation for activity.

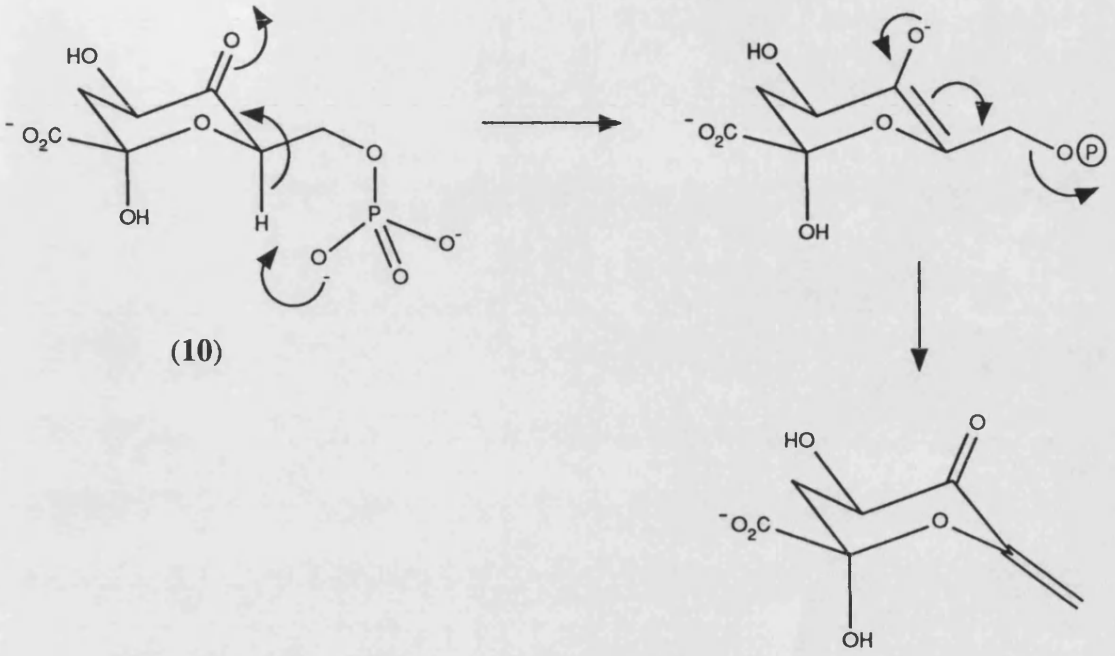
The cation Co^{2+} has been used in mechanistic studies, but Zn^{2+} is more likely required *in vivo*. The proposed mechanism (**Scheme 1.4**), involves an oxidation at C-5 which acidifies the C-6 proton and facilitates the elimination of phosphate. Subsequent reduction of the ketone gives the enol pyranose (**11**), which ring opens and undergoes an intramolecular aldol reaction affording 3-dehydroquinate (**5**).



Scheme 1.4 Proposed Mechanism for 3-Dehydroquinate Synthase.

This is a remarkably complex sequence of reactions for what is a relatively small, monomeric enzyme. Studies by Knowles[9] have suggested that the enzyme is not directly responsible for the β-elimination of phosphate. Instead, it is argued that

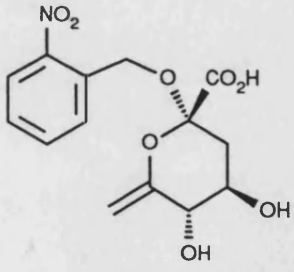
the substrate (**10**) is bound to the enzyme in a suitable conformation, such that one of the phosphoryl group oxygens is appropriately positioned to abstract the C-6 proton. Elimination then follows inexorably from the lability of the C-6 proton, due to substrate oxidation at C-5 (**Scheme 1.5**).



(**10**)

Scheme 1.5

Bartlett has synthesised the intermediate (**11**) by photochemical removal of the *O*-nitrobenzyl group from (**12**) as the final deprotection step.[10]

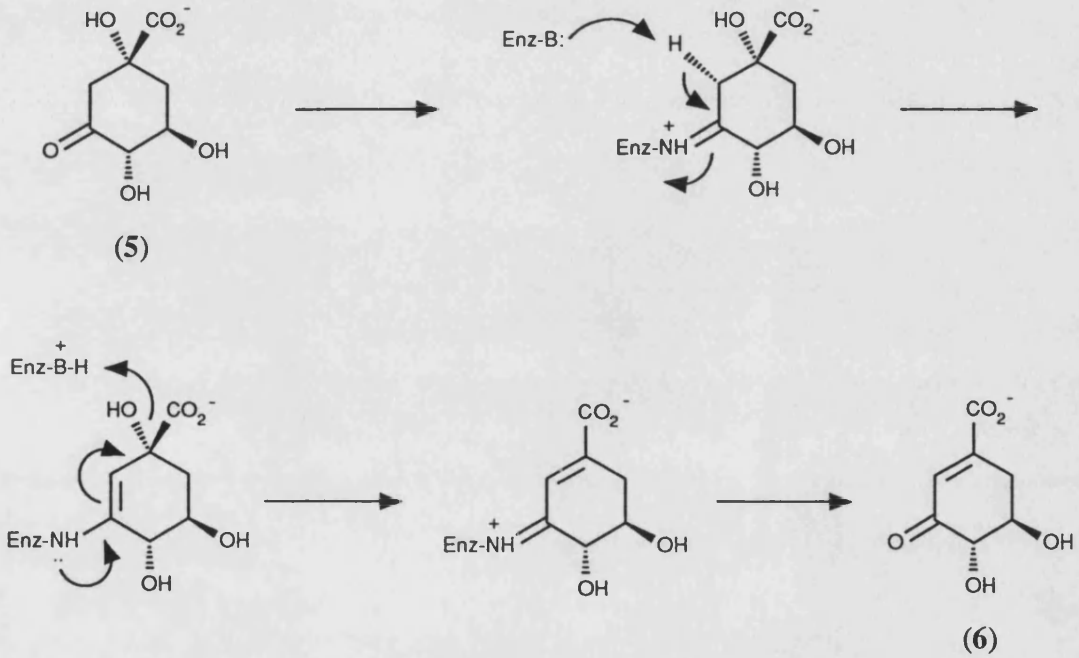


(**12**)

Photolysis of (**12**) in neutral aqueous solution effected a rapid, quantitative conversion to dehydroquinate (**5**). These results suggest that additionally, the

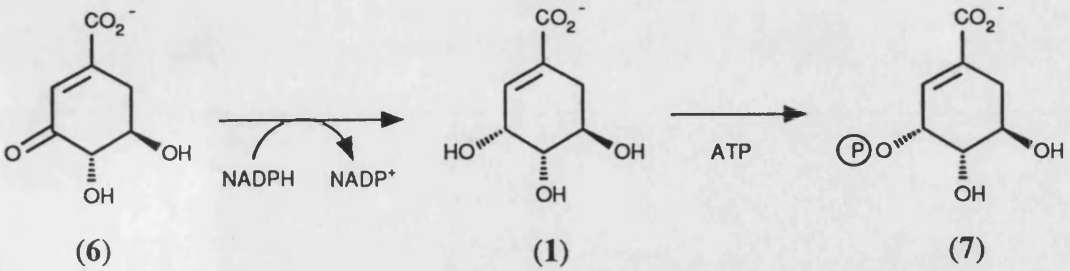
enzyme is not responsible for the last two steps of **Scheme 1.4**. Rather, dehydroquinate synthase produces the enol pyranose (**11**), which then rapidly and stereospecifically rearranges outside the active site to give 3-dehydroquinate (**5**).

The dehydration of 3-dehydroquinate (**5**) to 3-dehydroshikimate (**6**) is catalysed by 3-dehydroquinase and proceeds stereospecifically in a *cis* fashion. The active site of the enzyme is known to contain a lysine residue which forms a Schiff base with the keto group of 3-dehydroquinate, the arrangement of catalytic groups within the active site dictating the overall stereochemical course (**Scheme 1.6**).



Scheme 1.6 Proposed Mechanism for 3-Dehydroquinase.

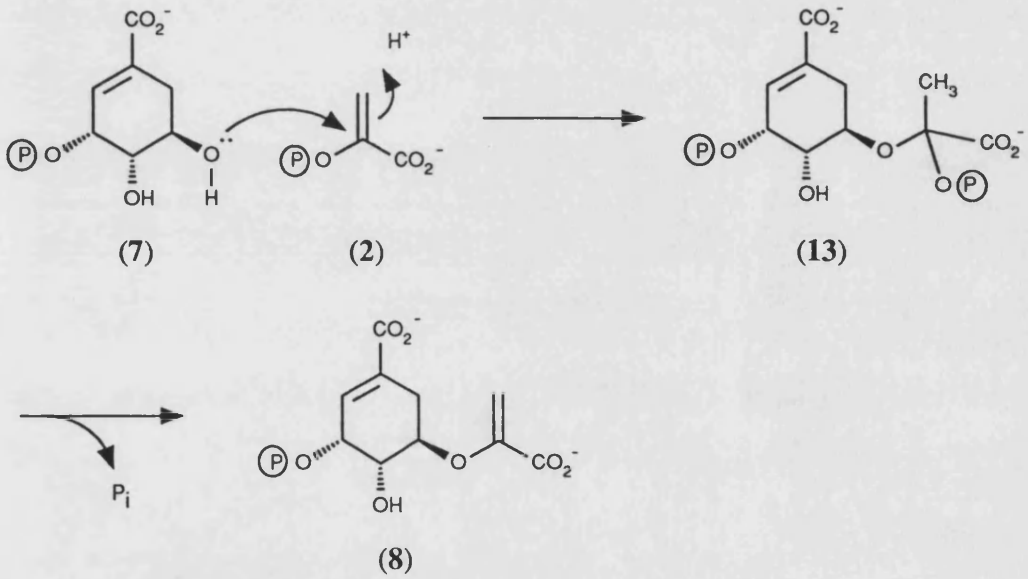
The next two steps in the common pathway are more straightforward. Shikimate dehydrogenase catalyses the reduction of dehydroshikimic acid, in the presence of NADPH, to give shikimic acid, which is then phosphorylated by shikimate kinase and ATP, to yield shikimic acid-3-phosphate (**7**) (**Scheme 1.7**).



(6) (1) (7)

Scheme 1.7

The formation of 5-enolpyruvylshikimate-3-phosphate (5-EPS-3-P) (**8**) constitutes the biochemically remarkable transfer of an intact enolpyruvyl group from PEP (**2**) to the 5-hydroxyl of (**7**), for which an addition-elimination sequence has been proposed (**Scheme 1.8**).[11]



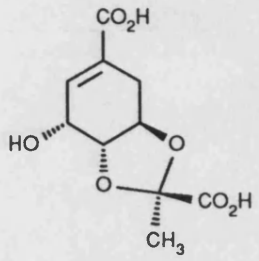
(7) (2) (13)

(8)

Scheme 1.8 Proposed Mechanism for 5-EPS-3-P Synthase.

The tetrahedral intermediate (**13**) has been isolated and characterised by Anderson.[12] Although stable under alkaline conditions, it hydrolyses readily at neutral pH and the configuration of the ketal carbon has not been elucidated. Further evidence for this mechanism was obtained by studying the enzyme catalysed reaction with ^{13}C labelled PEP and NMR resonances due to the enzyme bound tetrahedral

intermediate were observed.[13] A separate study by Evans,[14] in which resonances due to an enzyme free tetrahedral intermediate were claimed, was later shown to be in error,[15] the signals observed being due to the novel shikimate ketal (**14**), subsequently isolated by Sammons.[16] This side product is formed at equilibrium, but is not part of the normal catalytic pathway.

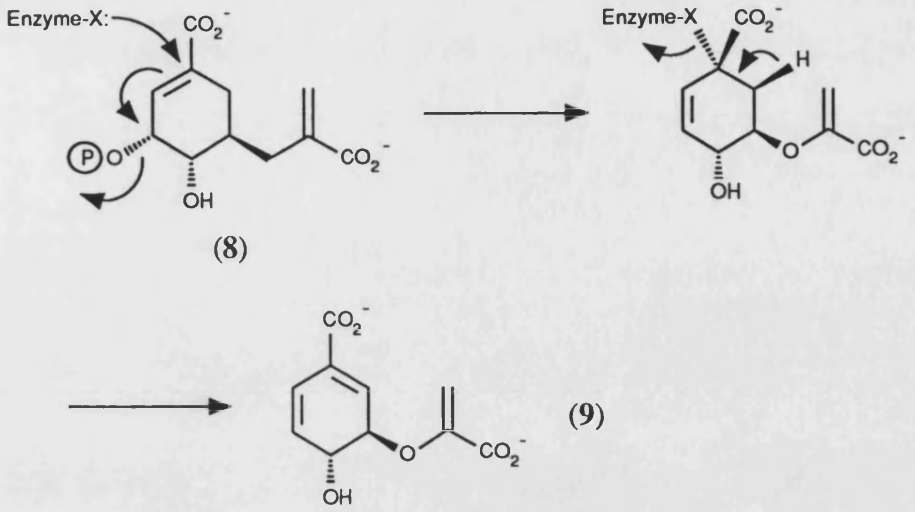


(**14**)

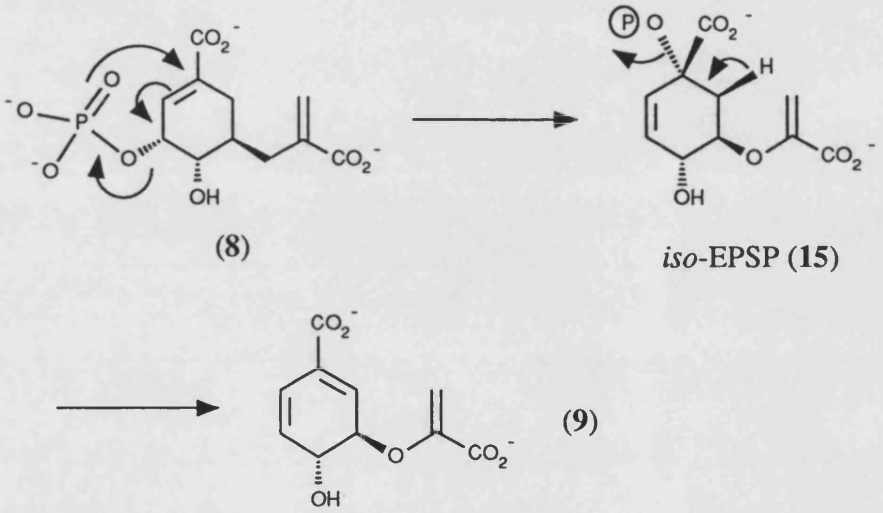
The final step of the common pathway involves the formation of chorismic acid (**9**) from 5-EPS-3-P (**8**), catalysed by chorismate synthase. Labelling experiments have shown that only the 6-*pro*-R hydrogen is lost and thus the overall transformation is a *trans*-1,4-elimination. Concerted 1,4-eliminations in cyclohexene systems proceed predominantly in a *cis* fashion, therefore, mechanisms involving a two stage process have been proposed. It is possible, however, that the enzyme causes the substrate to adopt a suitable conformation, such that a concerted *trans*-1,4-elimination is possible.

Floss[17] has speculated that a two-stage mechanism, in which an 'X-group' on the enzyme participates, might account for the overall *trans*-elimination (**Scheme 1.9**).

An interesting alternative proposed by Ganem[1d] involves a suprafacial 3,3-rearrangement of (**8**) to the allylic isomer, *iso*-EPSP (**15**), followed by *trans*-1,2-elimination (**Scheme 1.10**). However, *iso*-EPSP was synthesised by Bartlett[18] and was shown not be converted to chorismate by chorismate synthase, thus suggesting that *iso*-EPSP is not an intermediate in the reaction pathway.



Scheme 1.9 Proposed 'X-group' Mechanism for Chorismate Synthase.



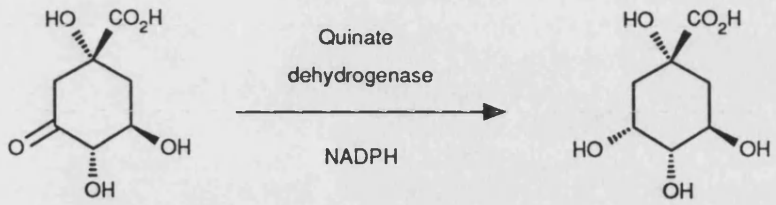
Scheme 1.10 Proposed Mechanism for Chorismate Synthase *via iso*-EPSP.

Chorismic acid is an unstable intermediate which can react in a variety of ways. From here the common pathway ends and the shikimate pathway branches out in several directions.

1.1.3 The Role of Quinic Acid

Quinic acid (**16**), widely found in the plant kingdom, is formed by an off-shoot of the common pathway. Once formed, by the reduction of

3-dehydroquinate (**5**) by NADPH, a reaction which is catalysed by quinate dehydrogenase (**Scheme 1.11**), it is not easily metabolised again. However, some micro-organisms are able to convert quinic acid into 3-dehydroquinate and thus metabolise the former as an alternate carbon source.



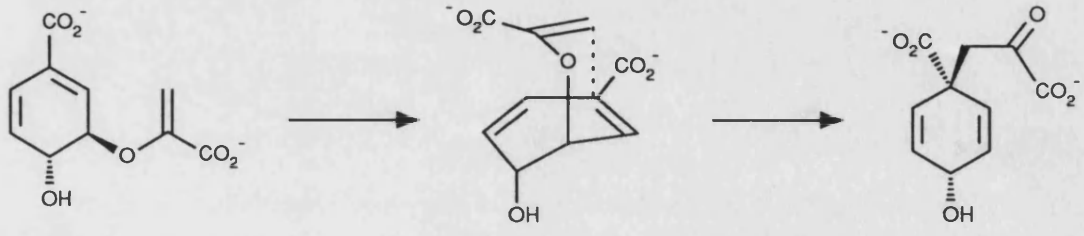
(**5**) (**16**)

Scheme 1.11

1.1.4 Branch Point Metabolism

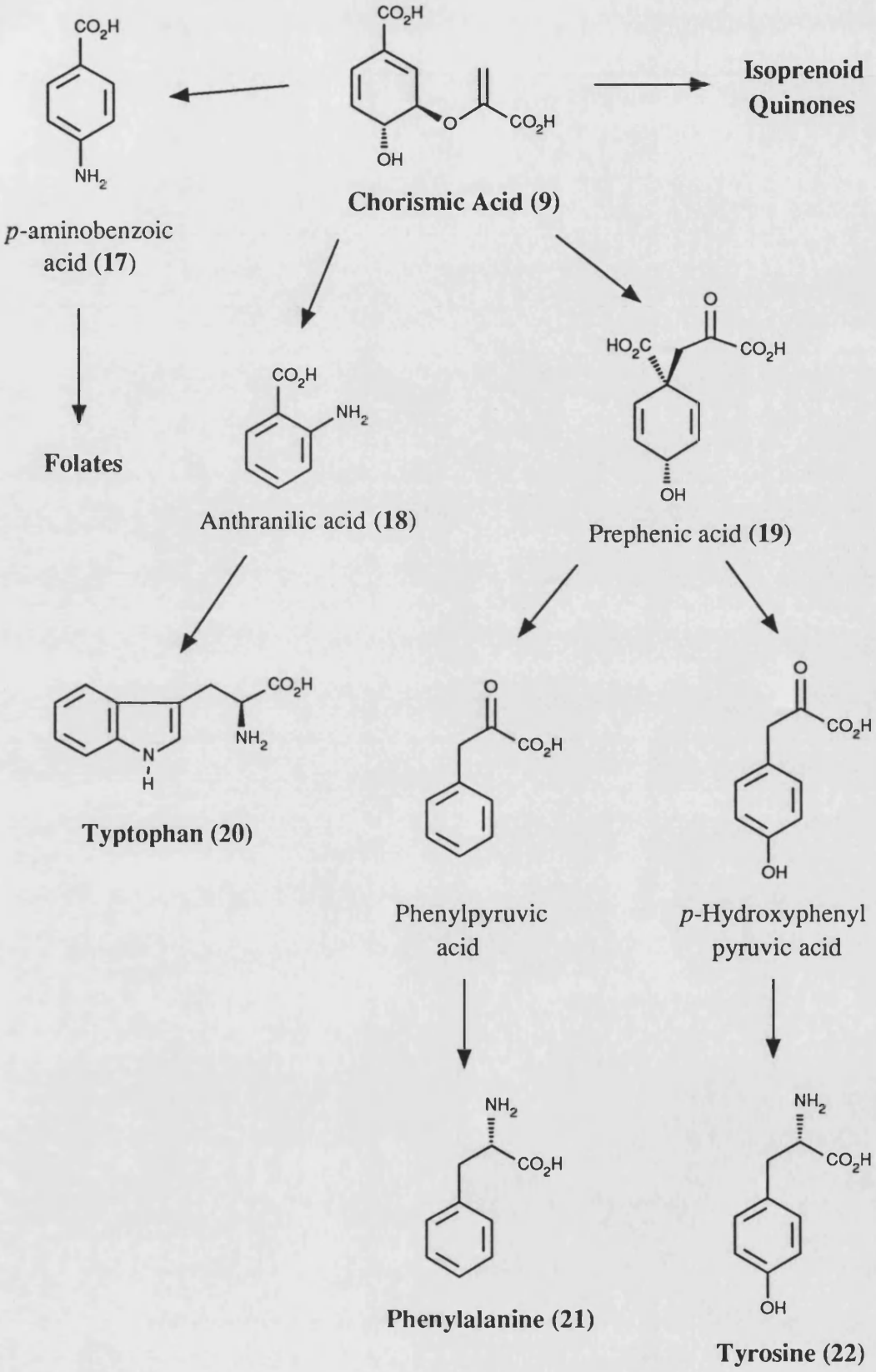
At chorismic acid the common pathway fragments into different routes, leading to the aromatic amino acids and diverse other compounds (**Scheme 1.12**). Amination of chorismic acid leads through anthranilic acid (**18**) to tryptophan (**20**). The other two aromatic amino acids phenylalanine (**21**) and tyrosine (**22**), are formed *via* rearrangement of chorismic acid to prephenic acid (**19**), in what is formally at least, a Claisen rearrangement. A unique reaction in biosynthesis, that has been proven to proceed through a chair-like transition state (**Scheme 1.13**).[19]

Another route from chorismate leads *via* *p*-aminobenzoic acid (**17**) to the folate group of coenzymes. The isoprenoid quinones which participate in electron transport and oxidative phosphorylation are also derived from chorismic acid.



(**9**) (**19**)

Scheme 1.13 Rearrangement of Chorismate by Chorismate Mutase.



Scheme 1.12 Biosynthesis of Aromatic Compounds from Chorismic Acid

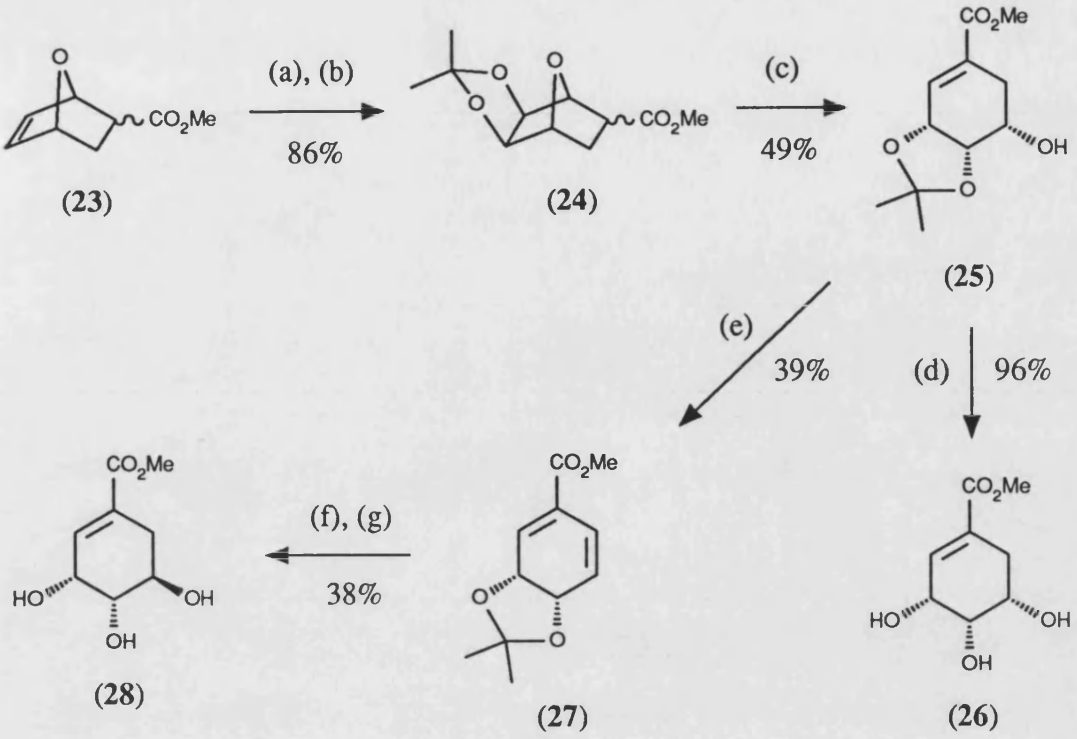
1.2 Syntheses of Shikimic acid

Chemists continue to be attracted to the stereospecific and chiral synthesis of shikimic acid and of structural variants, which may exhibit useful biological activity. The first total synthesis of shikimic acid was published in 1960 by Raphael *et al.*[20] Since then, many different approaches to both racemic and optically active forms of shikimic acid have been reported, which have recently been reviewed.[21] In this section the more recent synthetic studies will be reviewed, covering the literature from 1984 to date.

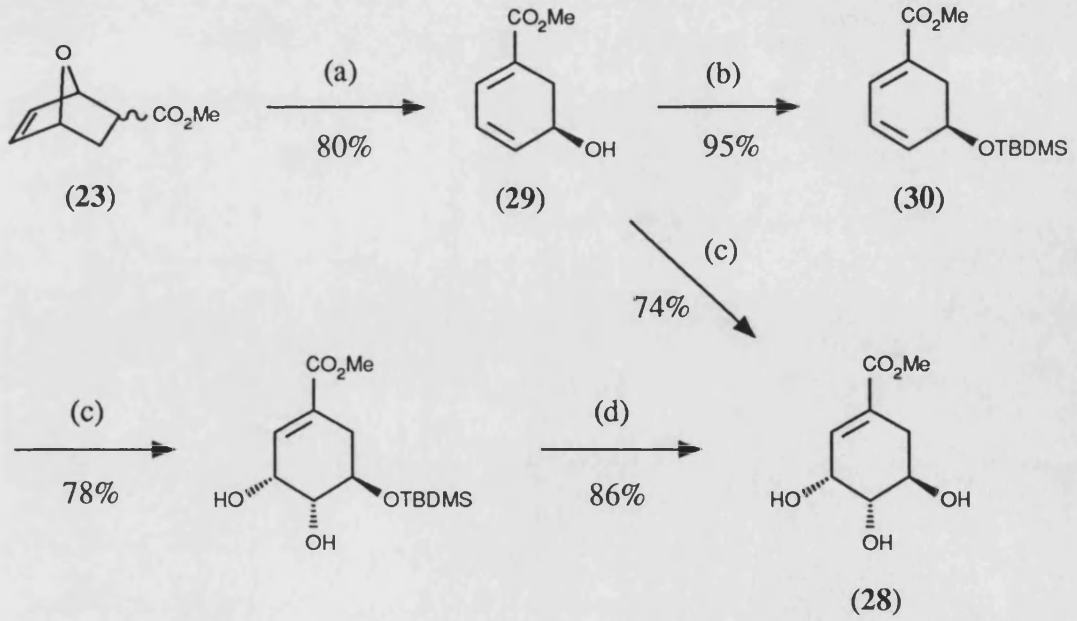
1.2.1 Campbell, Sainsbury *et al.*

Campbell, Sainsbury *et al.* have published a number of brief syntheses of racemic methyl shikimate (**28**) and methyl 5-*epi*-shikimate (**26**).[22] The starting material was the Diels-Alder adduct (**23**) of furan and methyl acrylate,[23] which was *cis*-hydroxylated and then protected as the acetonide (**24**) (**Scheme 1.14**). Base mediated opening of the oxabicyclo ring led to the acetonide of methyl 5-*epi*-shikimate (**25**), which was deprotected to afford (**26**). Direct attempts to invert the stereochemistry at C-5 of (**25**) were unsuccessful, however, dehydration under Mitsunobu[24] conditions yielded the diene (**27**), which on hydroboration, oxidation and deprotection gave methyl shikimate (**28**).

A more concise route was developed by reversing the order of the *cis*-hydroxylation and ring opening reactions (**Scheme 1.15**). Thus, initial ring opening of the adduct (**23**) afforded the diene (**29**), which on *cis*-hydroxylation yielded methyl shikimate (**28**) and methyl 5-*epi*-shikimate (**26**) in a 5:1 ratio. The stereoselectivity was improved by first protecting the hydroxyl of (**29**) as the TBDMS ether (**30**), which on osmylation and deprotection, gave (**28**) in 26% overall yield from the adduct (**23**).

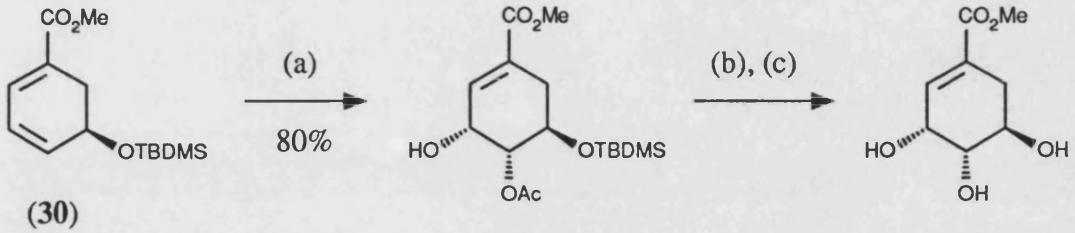


Scheme 1.14 Reagents: (a) OsO_4, H_2O_2, Me_2CO; (b) $(MeO)_2CMe_2$, *p*-TSA, Me_2CO; (c) $LiN(TMS)_2$, THF, -78°C; (d) aq. AcOH, 55°C; (e) DEAD, Ph_3P, THF; (f) B_2H_6, THF, H_2O_2; (g) Dowex W X-8 Resin (H^+), MeOH.



Scheme 1.15 Reagents: (a) $LiN(TMS)_2$, THF, -78°C; (b) TBDMSOTf, 2,6-lutidine, CH_2Cl_2, 0°C; (c) OsO_4, H_2O_2, Me_2CO; (d) *n*-Bu_4NF, THF, 0°C.

A further variation[25] employed a 'wet' Prévost reaction on the diene (**30**) to afford the hydroxy-acetate (**31**) (**Scheme 1.16**). Hydrolysis and deprotection gave rise to methyl shikimate in a similar overall yield (26%) from the adducts (**23**), but avoiding the use of the highly toxic osmium tetroxide.

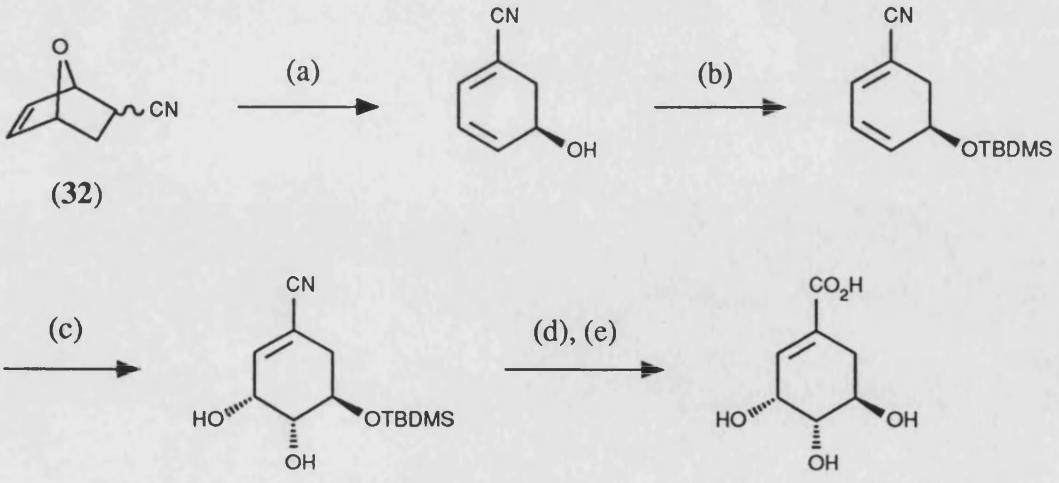


(**30**) (**31**) (**28**)

Scheme 1.16 Reagents: (a) AgOAc, I_2, AcOH, H_2O, 70°C; (b) aq. NH_3, MeOH; (c) *n*-Bu_4NF, THF, 0°C.

1.2.2 Rodrigo *et al.*

A synthesis by Rodrigo *et al.*,[26] published in the same year as the Campbell-Sainsbury route, detailed a similar approach to racemic shikimic acid, but utilised the adduct (**32**) of acrylonitrile and furan (**Scheme 1.17**).



(**32**) (**1**)

Scheme 1.17 Reagents: (a) LDA, THF, -78°C; (b) Protection; (c) OsO_4, Py; (d) *n*-Bu_4NF, THF; (e) OH^-, H_2O.

The overall yield from (**32**) was 32% and these workers also duplicated the Bath route from the Diels-Alder adduct (**23**) of furan and methyl acrylate.

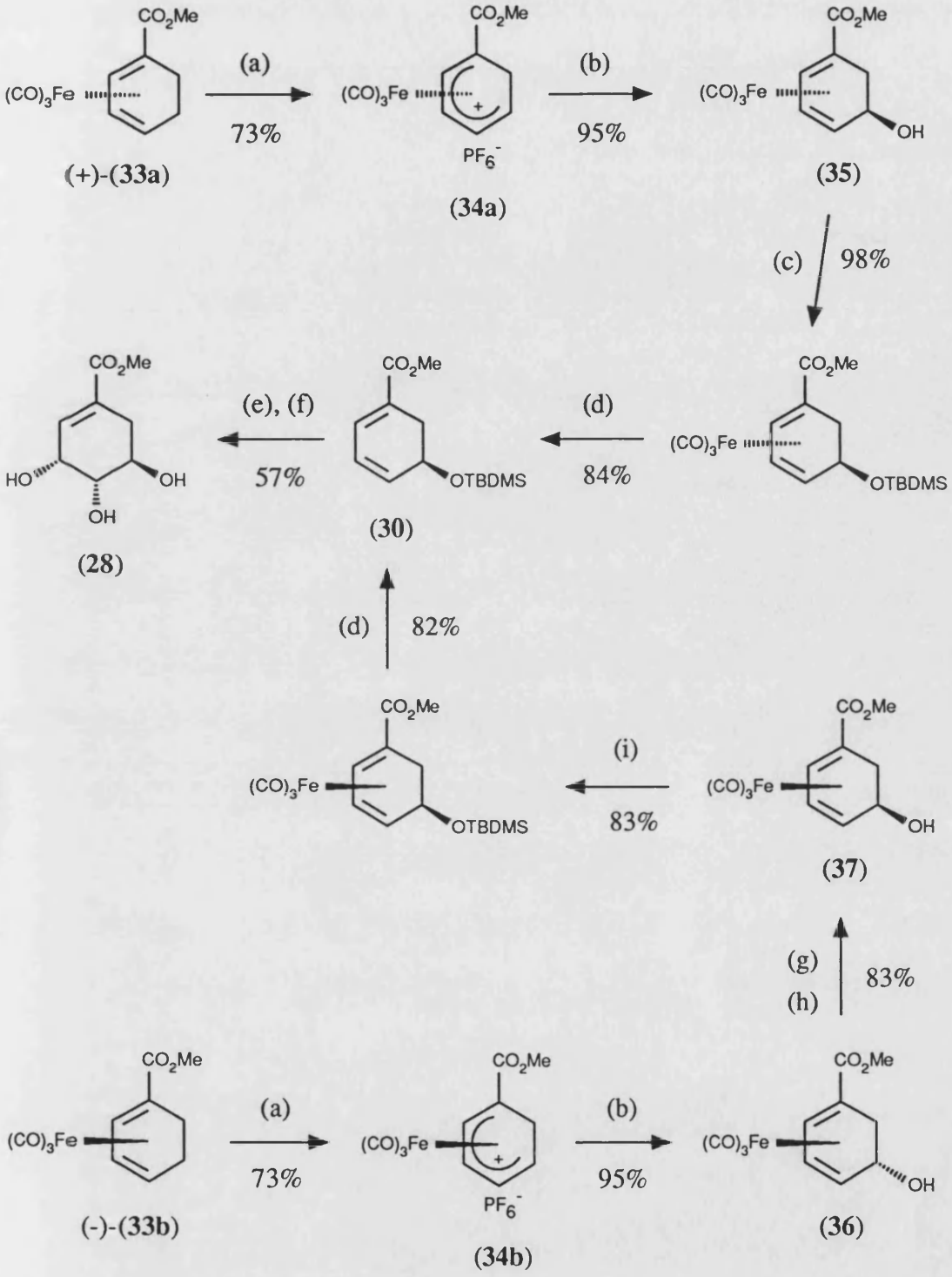
1.2.3 Birch *et al.*

The optically active form of the key intermediate (**30**), used in the synthesis of Campbell and Sainsbury (**Section 1.2.1**), was prepared by Birch *et al.*,[27] using iron tricarbonyl as a lateral control group. (-)-Methyl shikimate was prepared from either of the resolved iron tricarbonyl complexes (**33**), obtained from 1,4-dihydrobenzoic acid and previously used in an enantiospecific synthesis of gabaculine.[28]

Starting from the (+) complex (**33a**) (**Scheme 1.18**), the cationic salt (**34a**) was formed, which had previously been shown to react with nucleophiles solely at the 5-*exo* position.[29] Thus, reaction of (**34a**) with aqueous sodium hydrogen carbonate afforded the alcohol complex (**35**). Protection as the TBDMS ether and decomplexation, yielded the (+) enantiomer of the Bath intermediate (**30**), which was converted into (-)-methyl shikimate *via cis*-hydroxylation and deprotection as before.

Starting from the (-) complex (**33b**), a similar procedure led to the alcohol complex (**36**), having the wrong configuration at C-5. Inversion was achieved by Jones oxidation to the carbonyl compound, followed by a stereospecific reduction using sodium borohydride and zinc chloride, directed by the complexing group, to afford the alcohol complex (**37**). Protection and decomplexation gave the required diene (**30**).

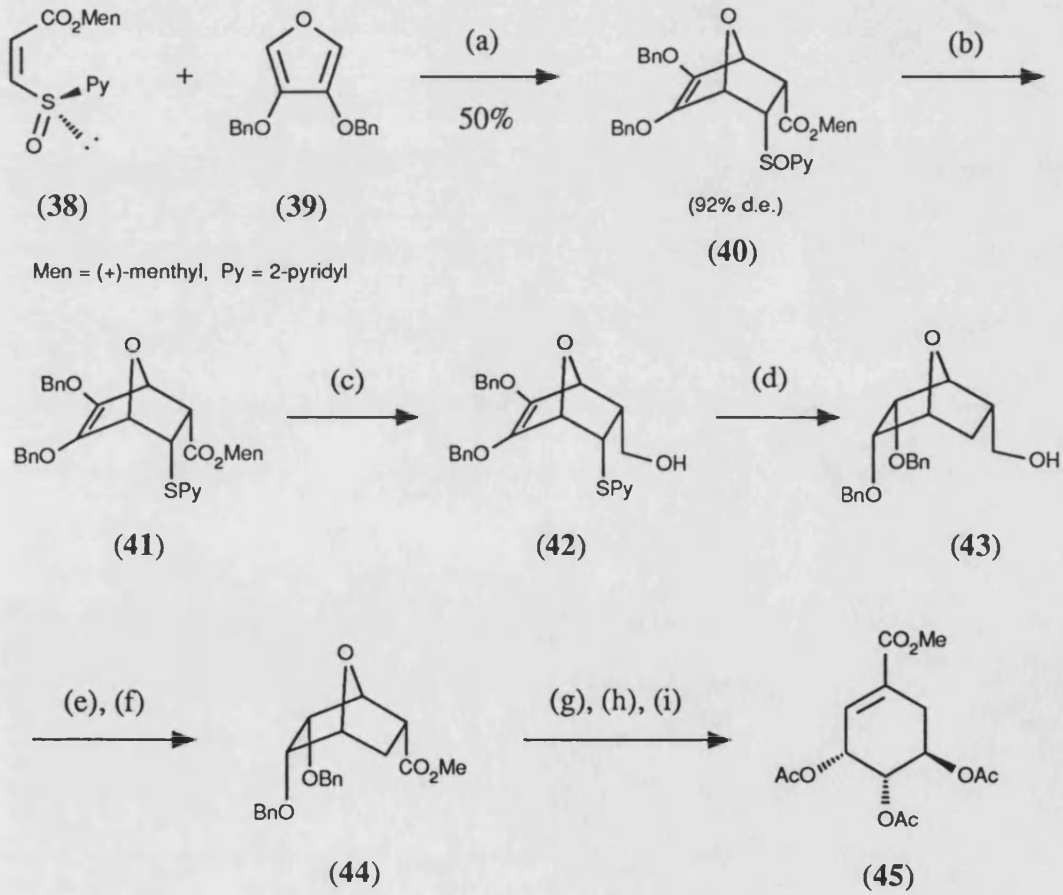
Therefore, although the initial complex (**33**) required resolution, both enantiomers could be used to give the same optically active product, of either absolute configuration.



Scheme 1.18 Reagents: (a) Ph_3C^+ PF_6^-, CH_2Cl_2; (b) $NaHCO_3$, MeCN, H_2O; (c) TBDMSCl, $(i\text{-Pr})_2NEt$, DMF; (d) Me_3NO, PhH; (e) OsO_4, Me_2CO; (f) $n\text{-Bu}_4NF$, THF; (g) CrO_3, Py, CH_2Cl_2; (h) $NaBH_4$, $ZnCl_2$, Et_2O; (i) TBDMSOTf, $(i\text{-Pr})_2NEt$, DMF.

1.2.4 Koizumi *et al.*

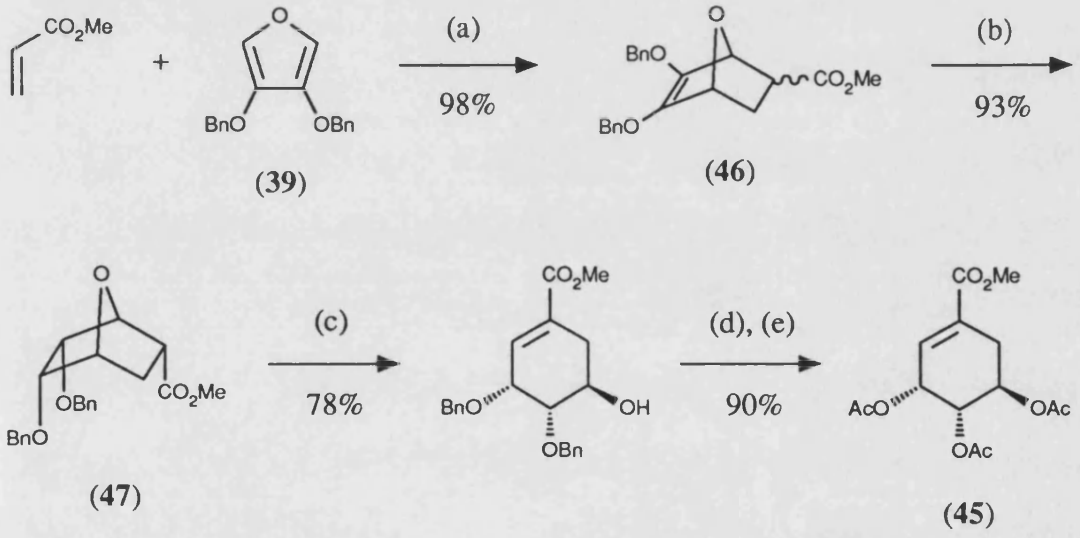
An enantioselective synthesis of methyl shikimate was reported by Koizumi *et al.*[30] An asymmetric Diels-Alder reaction of menthyl $(S)_s$-3-(2-pyridyl-sulphinyl)acrylate (**38**)[31] with 3,4-dibenzyloxyfuran (**39**)[32] (**Scheme 1.19**), yielded a mixture of *exo* and *endo* adducts, from which the major *endo* adduct (**40**) could be separated. Reduction of (**40**) to the sulphide (**41**), followed by reduction of the ester gave the alcohol (**42**). Treatment with Raney-nickel resulted in desulphurisation and hydrogenation of the double bond to give the *endo-cis*-dibenzyloxy derivative (**43**). Oxidation and esterification yielded the methyl ester (**44**), which upon ring opening, debenzylation and acetylation afforded (-)-methyl triacetylshikimate (**45**).



Scheme 1.19 Reagents: (a) Et_2AlCl; (b) PBr_3, DMF, 0°C; (c) $LiAlH_4$, Et_2O; (d) Raney-Ni, EtOH; (e) CrO_3, Py, Me_2CO; (f) CH_2N_2, MeOH, Et_2O; (g) $LiN(TMS)_2$, THF, -78°C; (h) TMSCl, NaI, MeCN; (i) Ac_2O, Py.

1.2.5 Koreeda *et al.*

The use of 3,4-dibenzyloxyfuran (**39**) as a precursor to shikimic acid, was also reported simultaneously by Koreeda *et al.*[33] in a synthesis of racemic methyl triacetylshikimate (**Scheme 1.20**). The Diels-Alder reaction of (**39**) with methyl acrylate, catalysed by zinc iodide, yielded the adduct (**46**) as a mixture of *exo* and *endo* adducts (ratio 15:1). The *endo* adduct was subsequently hydrogenated to afford the required *endo-cis*-dibenzyloxy derivative (**47**). Ring opening, debenzylation and subsequent purification of the resulting triol, as its triacetate, afforded methyl triacetylshikimate (**45**) in 60% overall yield from (**39**).

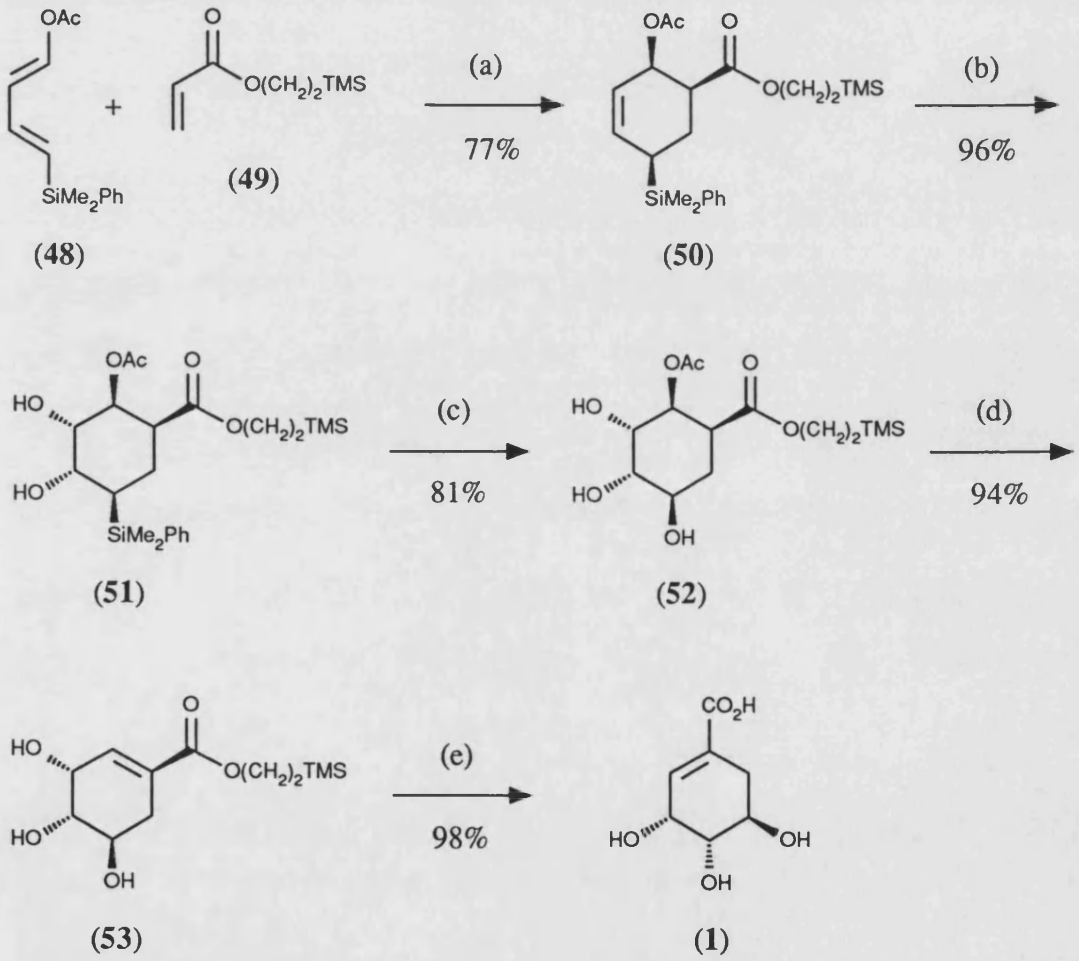


Scheme 1.20 Reagents: (a) ZnI_2; (b) H_2, PtO_2, EtOAc; (c) $LiN(TMS)_2$, THF, -78°C; (d) $BF_3.OEt_2$, EtSH, CH_2Cl_2, 0°C; (e) Ac_2O, Py.

1.2.6 Koreeda *et al.*

Koreeda has also recently published a concise total synthesis of racemic shikimic acid from (1*E*, 3*E*)-4-acetoxy-1-phenyldimethylsilyl-1,3-butadiene (**48**).[34] This was essentially an improved version of his earlier synthesis[35] and demonstrates the use of the diene (**48**) as a surrogate for (1*E*, 3*E*)-1,4-diacetoxy-1,3-butadiene (**Scheme 1.21**). The Diels-Alder reaction of (**48**) with 2-(trimethylsilyl)ethyl

acrylate (**49**) yielded the adduct (**50**) as the major product. *cis*-Hydroxylation afforded the diol (**51**), which was subjected to Flemming's one-pot buffered oxidation procedure[36] to yield the triol (**52**). Base mediated elimination produced 2-(trimethylsilyl)ethyl shikimate (**53**), which was deprotected to yield shikimic acid in 55% overall yield from (**48**). The use of the 2-(trimethylsilyl)ethyl ester avoided the problematic hydrolysis of methyl shikimate, which often results in aromatisation and has therefore been omitted from many syntheses.

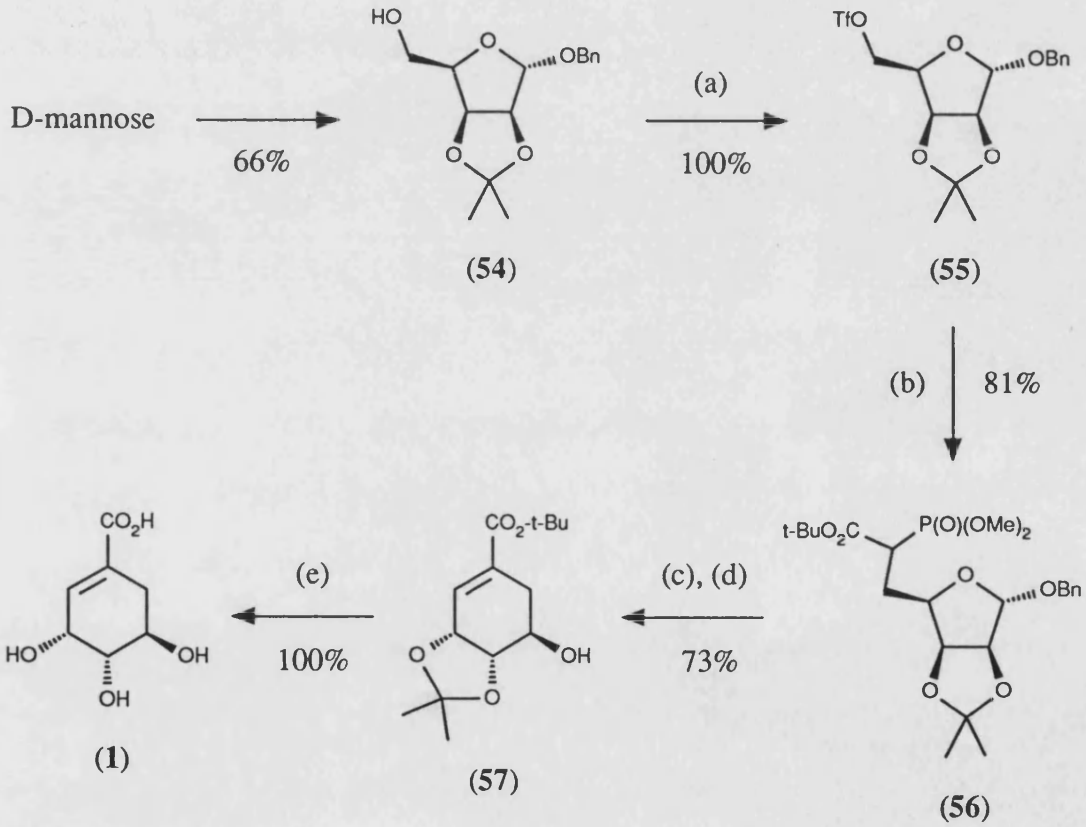


Scheme 1.21 Reagents: (a) hydroquinone monomethyl ester (cat.), xylenes, reflux; (b) OsO_4, NMO, THF, H_2O; (c) KBr, AcOOH, AcOH, AcONa; (d) DBU, THF; (e) *n*-Bu_4NF, THF.

1.2.7 Fleet *et al.*

Shikimic acid has been synthesised from carbohydrates[37] and also from quinic acid.[38] However, by far the most elegant approach starting from an optically active natural product, is that due to Fleet *et al.*[39]

Benzyl 2,3-*O*-isopropylidene-α-D-lyxofuranoside (**54**), obtained from D-mannose,[40] was converted to the triflate (**55**). Reaction with the sodium salt of *tert*-butyldimethoxyphosphorylacetate gave (**56**). On debenzylation and treatment with base, (**56**) underwent an intramolecular Wadsworth-Emmons olefination to afford *tert*-butyl 3,4-isopropylideneshikimate (**57**). Deprotection yielded (-)-shikimic acid in 39% overall yield from D-mannose.

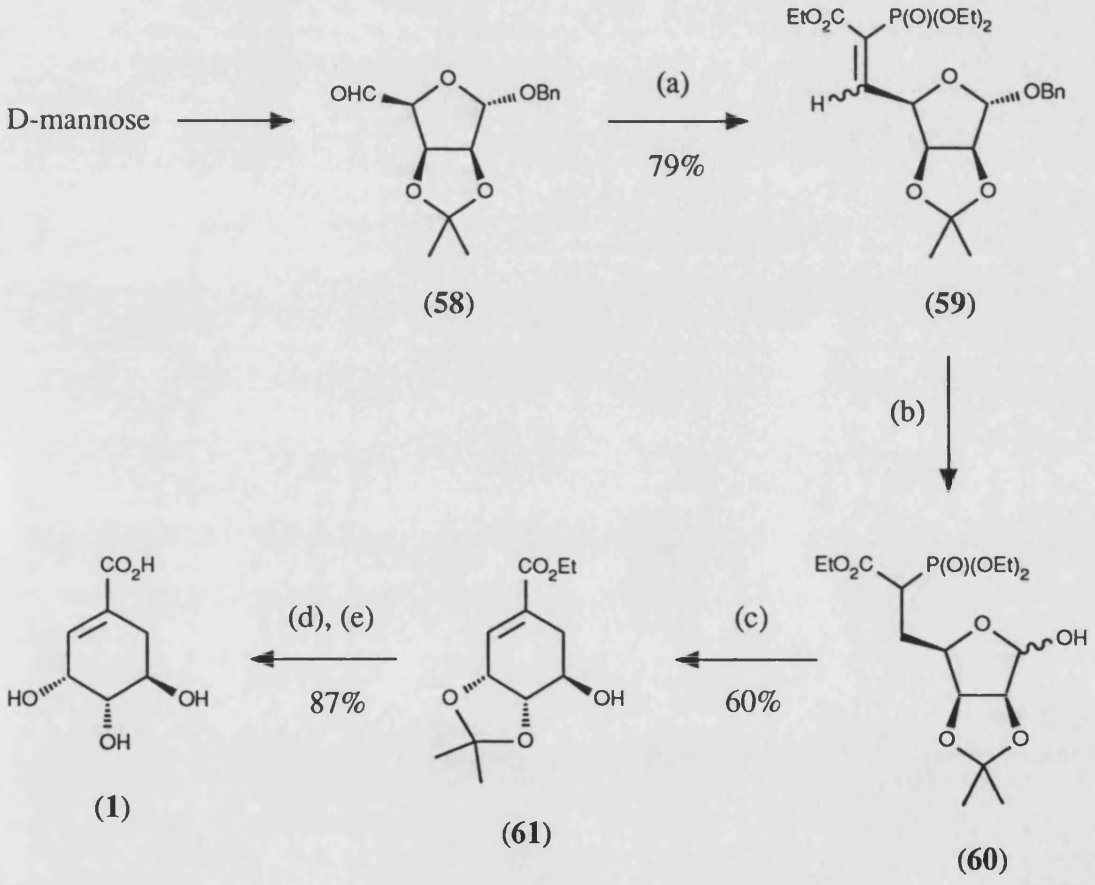


Scheme 1.22 Reagents: (a) Tf_2O, Py; (b) $[(MeO)_2P(O)CHCO_2{}^tBu]^-$ Na^+, DMF, 15-crown-5; (c) H_2, Pd-C; (d) NaH; (e) 60% aq. TFA.

1.2.8 Mirza *et al.*

An intramolecular olefination was also employed in the recent synthesis reported by Mirza *et al.*[41] Indeed, this synthesis is very similar to that of Fleet *et al.*

D-Mannose was converted into the suitably protected D-lyxose-5-aldehyde (**58**),[40] which was condensed with triethylphosphonoacetic acid to afford (**59**). Hydrogenation gave the hemiacetals (**60**), which on treatment with base underwent an intramolecular olefination to yield ethyl 4,5-isopropylideneshikimate (**61**). Deprotection afforded (-)-shikimic acid in 27% overall yield from D-mannose.



Scheme 1.23 Reagents: (a) $(EtO)_2P(O)CH_2CO_2Et$, *N*-methylmorpholine, $TiCl_4$, CCl_4, THF; (b) H_2, Pd-C, EtOH; (c) NaOEt, EtOH; (d) aq. NaOH, EtOH; (e) Dowex 50W-X4 (H^+), H_2O.

1.3 Synthesis of Later Intermediates in the Shikimate Pathway

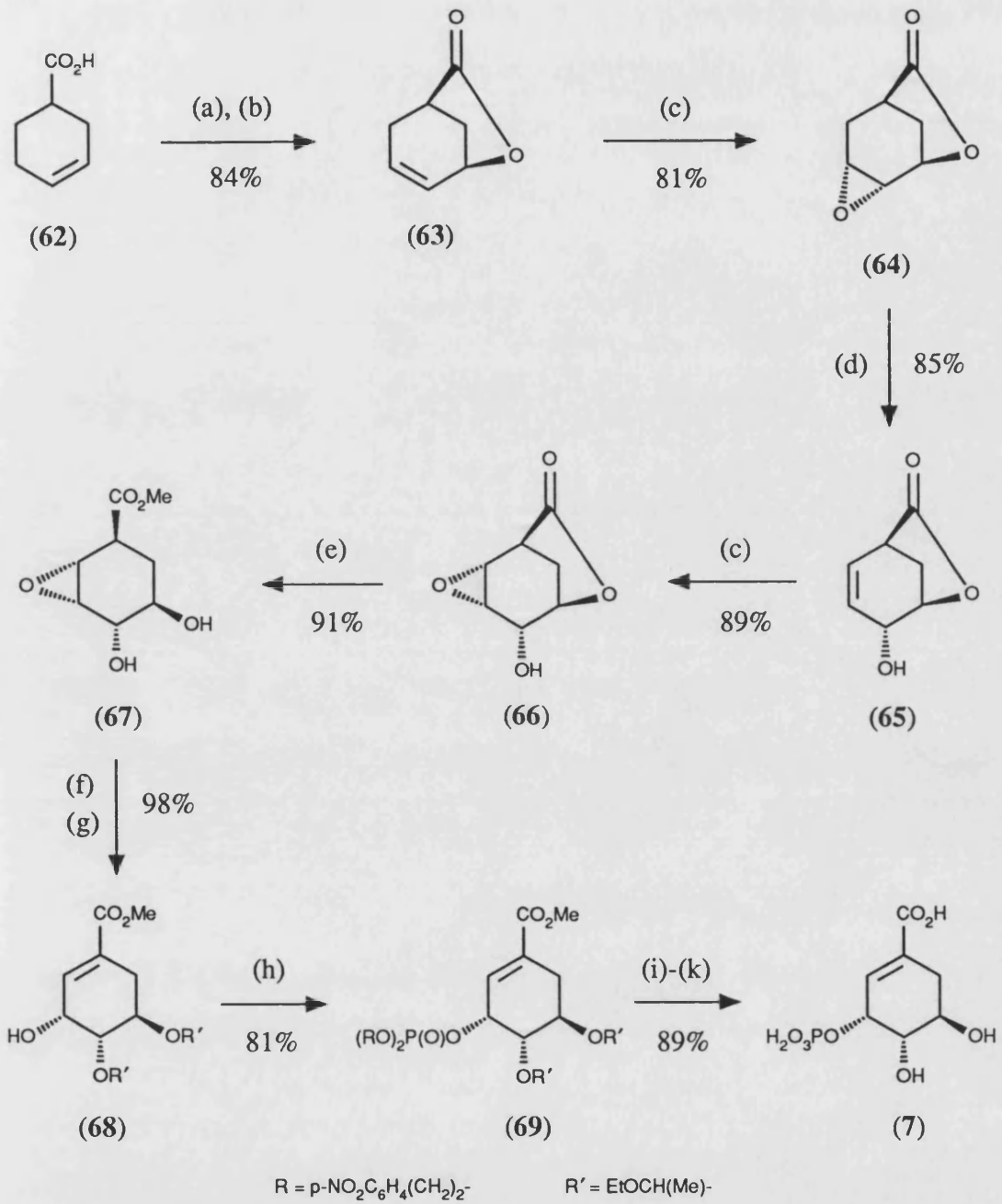
1.3.1 Shikimic Acid 3-Phosphate

The only reported total synthesis of shikimic acid 3-phosphate, is that described by Bartlett *et al.*,[42] in a total synthesis of shikimic acid (**Scheme 1.24**). Iodo-lactonisation of cyclohex-3-ene-1-carboxylic acid (**62**), followed by DBU induced elimination, afforded the lactone (**63**), which was converted to the epoxide (**64**). Opening of this epoxide with trimethylsilyl bromide, followed by elimination of the trimethylsilyl bromohydrin with DBU and aqueous workup gave the alcohol (**65**). Epoxidation afforded the epoxy alcohol (**66**), which upon methanolysis, yielded (±)-methyl shikimate in 46% overall yield from (**62**). The former had previously been prepared by Ganem[43] in another synthesis of shikimic acid.

Controlled methanolysis of (**66**) at a lower temperature gave the epoxy diol (**67**). Protection as the bis(ethoxyethyl) ether and opening of the epoxide, afforded the selectively protected shikimate derivative (**68**), in which the 3-hydroxyl is free. Phosphorylation was accomplished by using bis(*p*-nitrophenylethyl) phosphoro-chloridate[44] to yield the phosphate triester (**69**). Deprotection of the phosphate by treatment with DBU, followed by hydrolysis and cleavage of the acetal protecting groups, yielded (±)-3-phosphoshikimic acid (**7**), which was purified by ion exchange chromatography of the sodium salt.

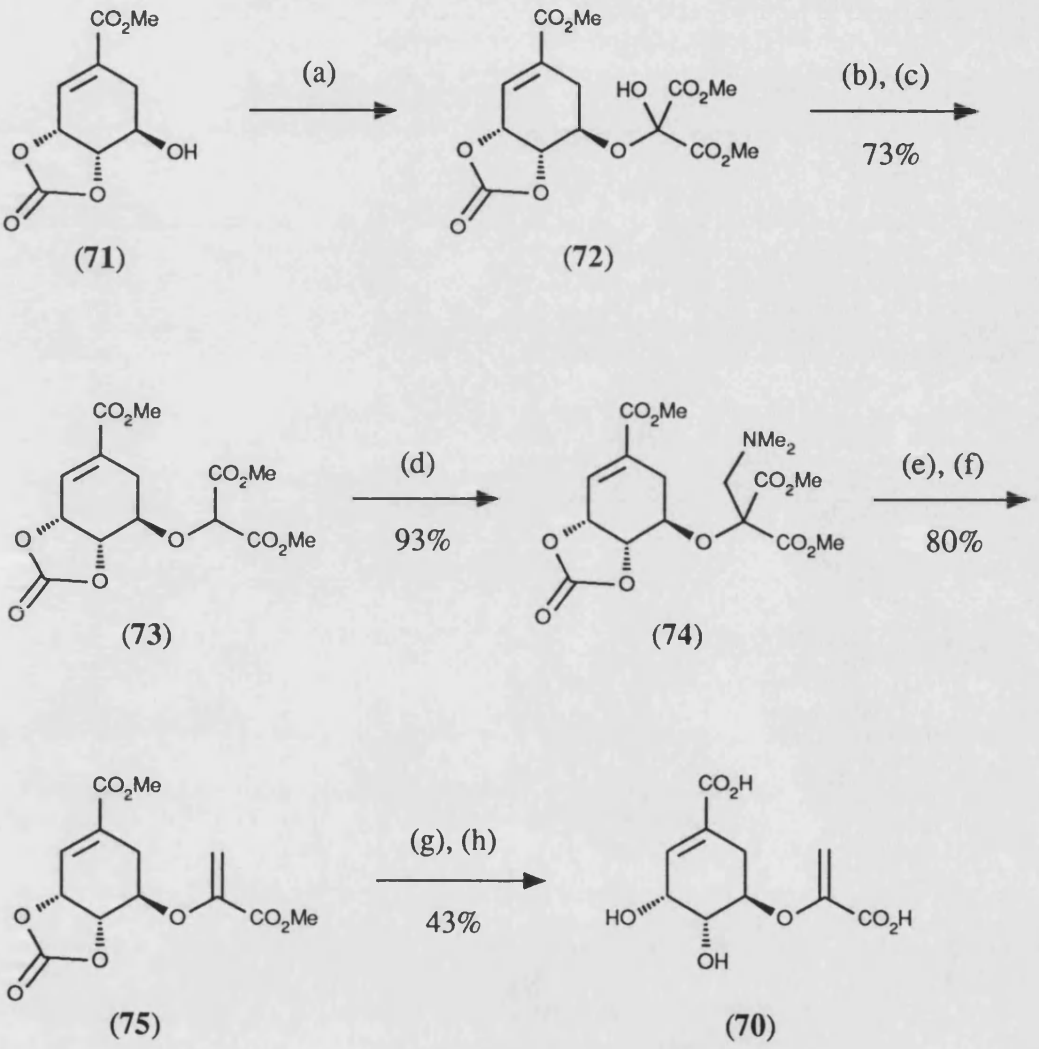
1.3.2 5-Enolpyruvylshikimate-3-phosphate (5-EPS-3P)

The construction of the enolpyruvyl functionality of 5-EPS-3-P and chorismic acid, was first demonstrated by Berchtold *et al.*[45] in a synthesis of 5-enolpyruvylshikimate (**70**) (**Scheme 1.25**). The carbonate derivative of (-)-methyl shikimate was reacted with dimethyloxomalonate to afford the hemiketal (**72**). Treatment with thionyl chloride followed by reduction, yielded (**73**), which was converted to the Mannich base (**74**). Quaternisation gave the quaternary ammonium



Scheme 1.24 Reagents: (a) I_2, KI, $NaHCO_3$, H_2O; (b) DBU, THF, reflux; (c) 3,5-dinitroperbenzoic acid, CH_2Cl_2, reflux; (d) TMSBr, Ph_3P, MeCN then DBU, reflux; (e) K_2CO_3, MeOH, 0°C; (f) ethyl vinyl ether, PPTS, THF; (g) K_2CO_3, MeOH; (h) bis(*p*-nitrophenylethyl) phosphorochloridate, Py, DMAP, CH_2Cl_2; (i) DBU, $CHCl_3$; (j) aq. NaOH; (k) Dowex 50W-X8 (H^+), H_2O.

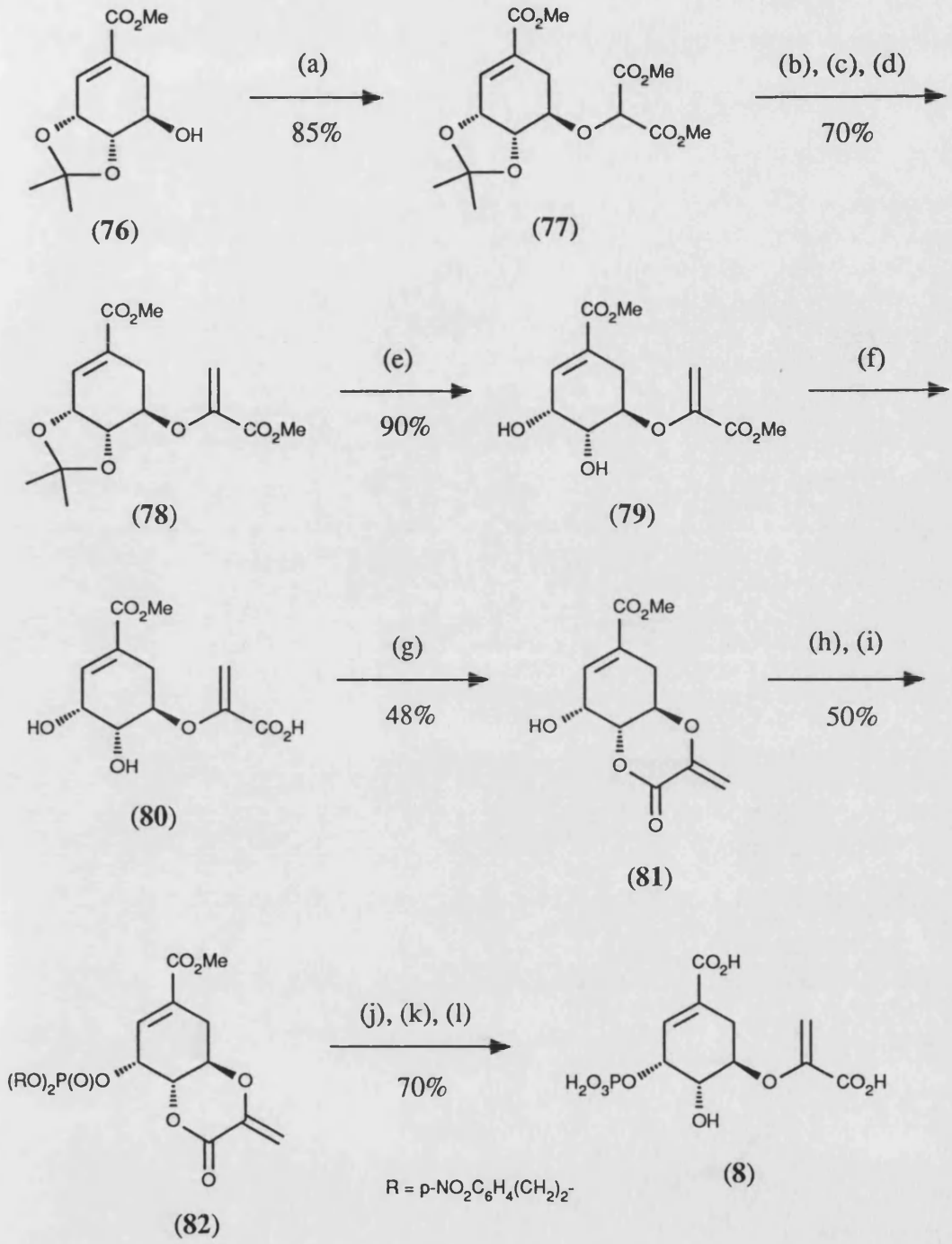
iodide, which upon heating underwent decarboxylation and elimination to afford (**75**). Hydrolysis of (**75**) gave 5-enolpyruvylshikimic acid (**70**) or 'compound Z_1'.[46] Compound Z_1 has been observed as a secondary metabolite from hydrolytic cleavage of the phosphate ester group of 5-EPS-3-P, but has no known biological function.



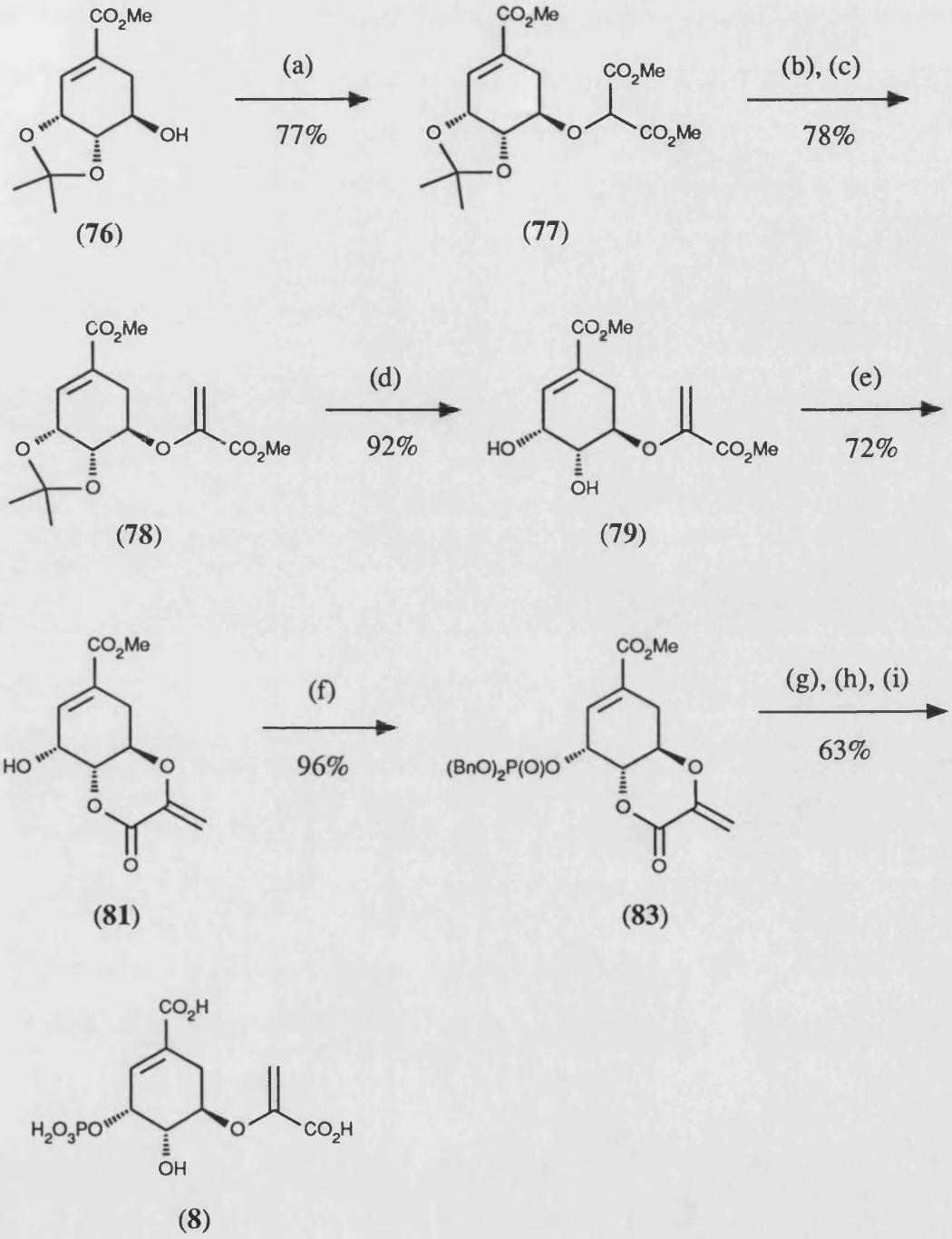
Scheme 1.25 Reagents: (a) $(MeO_2C)_2CO$, PhH, reflux; (b) $SOCl_2$, Py, THF, 0°C; (c) Zn, 90% aq. AcOH, 0°C; (d) $CH_2{=}NMe_2^+\ I^-$, Et_3N, CH_2Cl_2; (e) MeI, CH_2Cl_2, reflux; (f) DMSO, 80°C; (g) aq. NaOH; (h) Amberlite IR 120 (+), H_2O.

The first synthesis of 5-EPS-3-P was reported by Ganem *et al.*[47] The acetonide of (-)-methyl shikimate (**76**) was converted to the alkoxymalonate (**77**), by the $Rh_2(OAc)_4$ catalysed insertion of dimethyl diazomalonate (**Scheme 1.26**). Reaction of (**77**) with Eschenmoser's reagent, quaternisation and decarboxylation/elimination, in a similar sequence to that used by Berchtold,[45] led to the enolpyruvate (**78**). Deprotection afforded the diol (**79**), which could be selectively hydrolysed to yield the monoacid (**80**). Cyclisation of (**80**) gave the bicyclic lactone (**81**) and thus allowed phosphorylation selectively at the C-3 hydroxyl group. This was achieved *via* the bis(*p*-nitrophenylethyl) phosphite, which was oxidised to the phosphate (**82**). The 'one-pot' deprotection of (**82**) furnished 5-EPS-3-P (**8**) as the tetrasodium salt after ion-exchange chromatography.

A similar synthesis was published by Bartlett *et al.*[48] (**Scheme 1.27**), in which the early steps are virtually identical. However, the bicyclic lactone (**81**) was produced by direct cyclisation of the diol (**79**) with potassium carbonate. Phosphorylation was achieved using tetrabenzylpyrophosphate to afford the phosphate triester (**83**). Cleavage of the benzyl esters, with trimethylsilyl bromide, followed by alkaline hydrolysis, gave 5-EPS-3-P, which was obtained as the tetrasodium salt after ion-exchange chromatography.



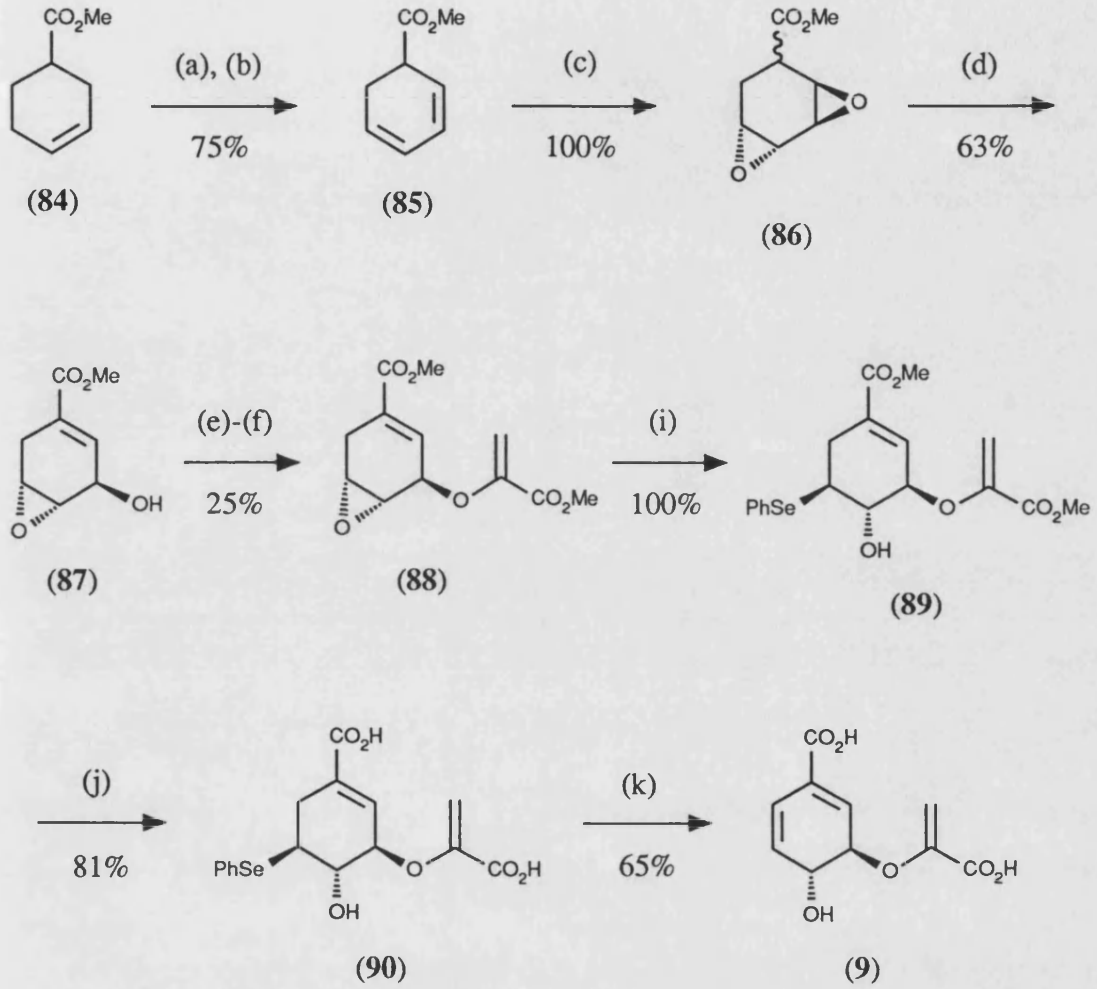
Scheme 1.26 Reagents: (a) $(MeO_2C)_2CN_2$, $Rh_2(OAc)_4$, PhH, reflux; (b) $CH_2{=}NMe_2^+$ I^-, Et_3N, CH_2Cl_2; (c) MeI, CH_2Cl_2; (d) DMSO, 95°C; (e) 80% aq. AcOH, 70°C; (f) aq. NaOH (1.1 equiv.), THF; (g) DCC, DMAP, THF; (h) PCl_3, Py, THF then *p*-$NO_2C_6H_4(CH_2)_2OH$; (i) I_2, H_2O, -78 to 0°C; (j) DBU, Py; (k) aq. NaOH; (l) Amberlite IR 120 (+).



Scheme 1.27 Reagents: (a) $(MeO_2C)_2CN_2$, $Rh_2(OAc)_4$, PhH, 85°C; (b) $CH_2{=}NMe_2^+ I^-$, Et_3N, CH_2Cl_2; (c) MeI, MeCN, reflux; (d) 65% aq. AcOH, THF, 70°C; (e) K_2CO_3, MeCN; (f) LDA, $[(BnO)_2P(O)]_2O$, THF, -78°C; (g) TMSBr, Py, CH_2Cl_2, 0°C; (h) aq. NaOH then ion-exchange resin.

1.3.3 Chorismic Acid

The total synthesis of (±)-chorismic acid was first reported in 1982 by Berchtold *et al.*[45,49] An improved synthesis was later published by the same group (**Scheme 1.28**).[50]

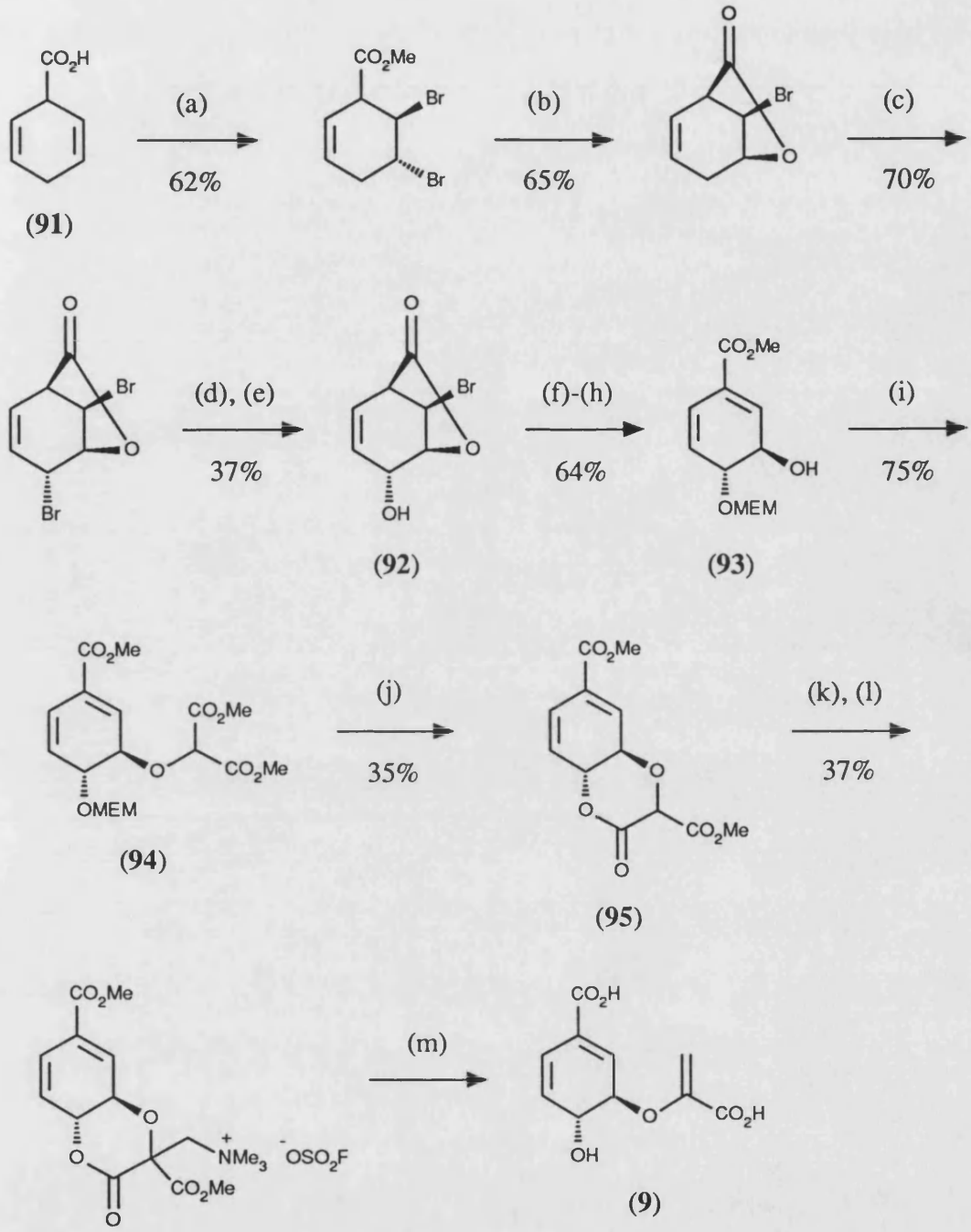


Scheme 1.28 Reagents: (a) NBS, AIBN, CCl_4, reflux; (b) *n*-Bu_3SnH, AIBN, PhH, reflux; (c) *m*-CPBA, CH_2Cl_2; (d) DBU, CH_2Cl_2; (e) $(MeO_2C)_2CN_2$, $Rh_2(OAc)_4$, PhH, 65°C; (f) $CH_2{=}NMe_2^+\ I^-$, Et_3N, CH_2Cl_2; (g) MeI, CH_2Cl_2; (h) DMSO, 80°C; (i) $(PhSe)_2$, $NaBH_4$, MeOH; (j) aq. NaOH, THF, 0°C; (k) H_2O_2, DMA, Me_2CO, -35 to 20 °C.

Bis allylic bromination of (**84**) gave a mixture of dibromides, that were

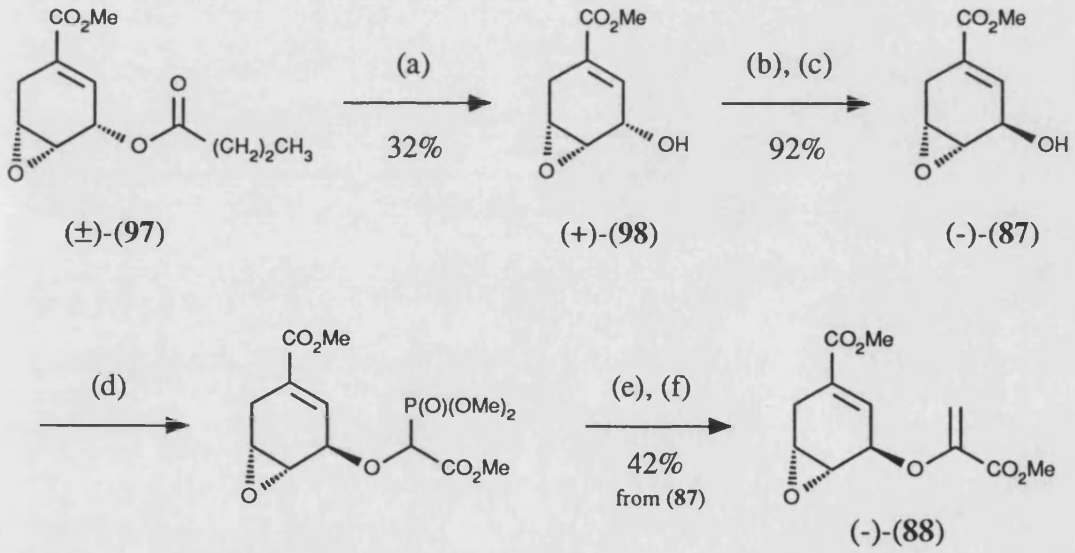
debrominated to afford the diene (85). Epoxidation of (85) yielded (86), which was isomerised to (87) on treatment with DBU. The enolpyruvyl side chain was constructed using either Ganem's[47] or Berchtold's own[45,49] procedure to afford (88). Opening of the epoxide with $PhSe^-$ gave (89), which was hydrolysed to the diacid (90). Selenoxide elimination from (90), in the presence of 3,5-dimethoxyaniline as a PhSeOH scavenger, yielded chorismic acid (9). The hydrolysis of (89) was carried out before selenoxide elimination, since hydrolysis of dimethyl chorismate gave poor yields of chorismic acid due to aromatisation.

A total synthesis of (±)-chorismic acid was also published in 1982 by Ganem *et al.*[51] The bicyclic allylic alcohol (92) was first prepared in five steps from 1,4-dihydrobenzoic acid (91)[52] (**Scheme 1.29**). Protection of the hydroxyl as its MEM ether, saponification and esterification yielded (93). Attachment of the enolpyruvyl functionality to (93) proved difficult, but was achieved in a stepwise manner as follows. Coupling of (93) with dimethyl diazomalonate, catalysed by $Rh_2(OAc)_4$, afforded (94), which was cyclised to give the bicyclic lactone (95). Alkylation of (95) with Eschenmoser's salt could not be achieved. However, Potier's salt was employed successfully, followed by quaternisation to yield (96). Finally the 'one-pot' hydrolysis, decarboxylation and β-elimination in aqueous sodium hydroxide afforded chorismic acid (9). Dimethyl chorismate could also be obtained from (96) by variation of reaction conditions.



Scheme 1.29 Reagents: (a) Br_2, CH_2Cl_2; (b) aq. $NaHCO_3$; (c) NBS, $(PhCO_2)_2$, CCl_4, reflux; (d) NaOAc, HMPA; (e) 10% aq. H_2SO_4, THF, reflux; (f) MEM-Et_3N^+ Cl^-, MeCN, reflux; (g) aq. KOH, THF; (h) MeI, HMPA; (i) $(MeO_2C)_2CN_2$, $Rh_2(OAc)_4$, PhH, 65°C; (j) *p*-TSA, PhH, H_2O; (k) $CH_2{=}NMe_2^+$ $CF_3CO_2^-$; (l) FSO_2OMe, $CDCl_3$; (m) aq. NaOH, THF.

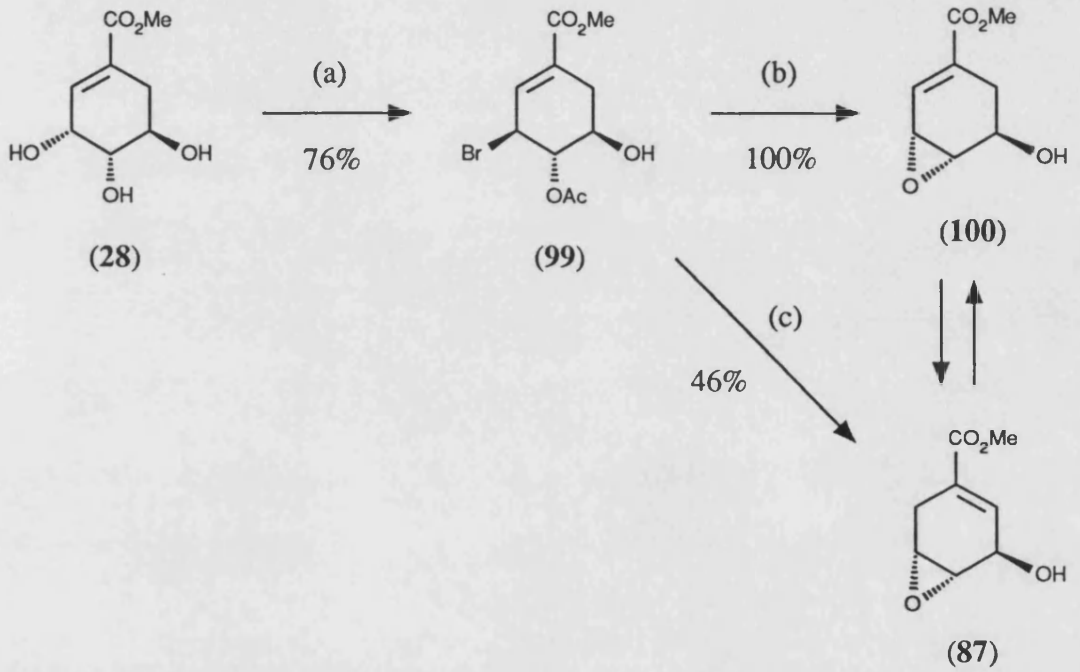
The enantiospecific synthesis of chorismic acid has been accomplished by both Berchtold and Ganem. The former has described the synthesis of an enantiomerically pure intermediate for his earliest synthesis of chorismic acid,[45,49] from quinic acid.[53] The key intermediate (**87**) used in his second synthesis (**Scheme 1.28**), was also prepared in chiral form.[54] An enantioselective enzymatic hydrolysis of the *n*-butyrate ester (**97**), followed by inversion of the configuration of the carbinol carbon of (+)-(**98**), yielded (-)-(**87**) (**Scheme 1.30**). The enolpyruvyl side chain was attached in a different manner, *via* coupling with methyl diazophosphonoacetate and reaction with formaldehyde to afford (-)-(**88**). Transformation of (-)-(**88**) to (-)-chorismic acid was accomplished as described previously.[50]



Scheme 1.30 Reagents: (a) cholesterol esterase, H_2O, pH 7.8, 0 to 5°C; (b) *i*-$PrO_2CN{=}NCO_2$-*i*-Pr, Ph_3P, AcOH, THF; (c) NaOMe, MeOH; (d) $MeO_2CC(N_2)P(O)(OMe)_2$, $Rh_2(n\text{-}C_7H_{15}CO_2)_4$, PhH, reflux; (e) $LiN(TMS)_2$, THF, -78°C; (f) H_2CO, -78°C.

An alternative approach to Berchtold's epoxide (-)-(**87**) was developed recently by Ganem *et al.*[55] The reaction of (-)-methyl shikimate (**28**) with

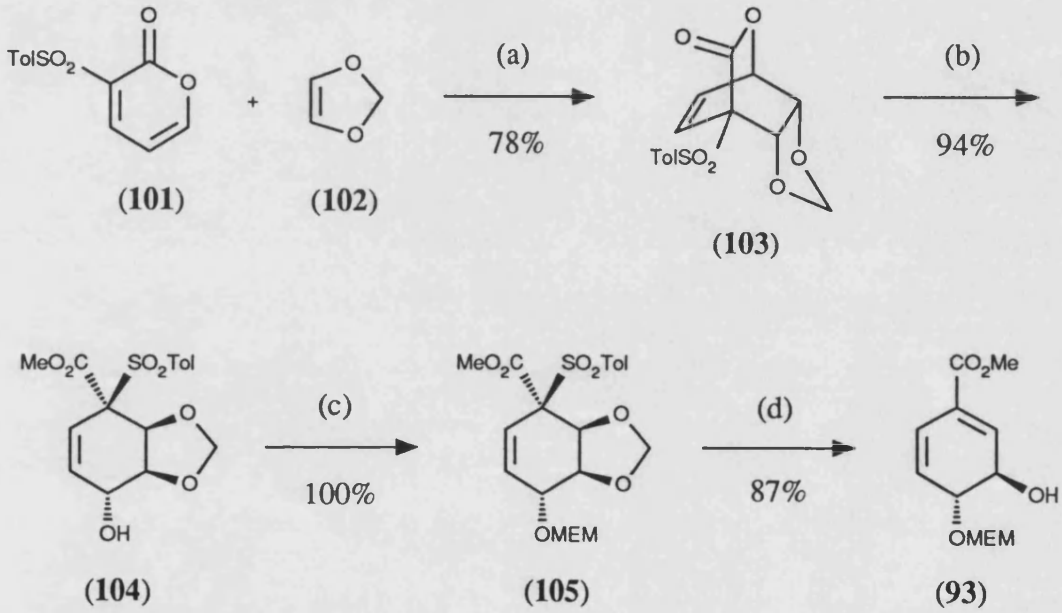
2-acetoxyisobutyryl bromide[56] furnished *trans*-bromoacetate (**99**) (**Scheme 1.31**). Transesterification of (**99**), with sodium methoxide in methanol, led to the epoxide (**100**). This epoxide, also known as methyl 3,4-anhydroshikimate, had previously been reported in the literature,[57] although the specific rotation was different from that observed by Ganem.[55] This discrepancy was attributed to a Payne rearrangement to (**87**), previously undetected by the earlier workers.[58] Prolonged exposure of (**100**) to sodium methoxide produced a 1:3 equilibrium mixture of (**100**):(**87**). The 'one-pot' conversion of the bromoacetate (**99**) to the epoxide (**87**) was possible, thus leading to a simple two step synthesis from (-)-methyl shikimate.



Scheme 1.31 Reagents: (a) α-acetoxyisobutyryl bromide, MeCN, 0°C; (b) NaOMe, MeOH, 0°C, 30 min; (c) NaOMe, MeOH, 0°C, 30 min then 50°C, 35 min.

A concise synthesis of Ganem's key intermediate (**93**)[51] has recently been published by Posner *et al.*[59] The cycloaddition of a 1,2-dioxygenated alkene (**102**) with a 3-sulphonyl-2-pyrone (**101**), yielded the bicyclic lactone (**103**) (**Scheme 1.32**). The presence of the electron withdrawing sulphonyl group on the 2-pyrone

and an electron rich alkene, facilitates this inverse-electron-demand Diels-Alder reaction under milder conditions. Thus cycloreversion, with loss of CO_2 and subsequent aromatisation, which occurs at more forcing conditions, is avoided. Methanolysis of (**103**) afforded the highly functionalised cyclohexene (**104**). Protection of the hydroxyl as the MEM ether (**105**), followed by reductive cleavage of the sulphonyl group, gave the mono-protected *trans*-diol (**93**), which had previously been converted to (±)-chorismic acid.



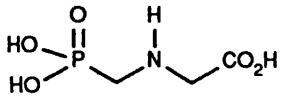
Scheme 1.32 Reagents: (a) 80°C, 36 h; (b) NaOMe, MeOH, 0°C; (c) *i*-Pr_2NEt, MEMCl, $CHCl_3$, reflux; (d) Zn, THF, sat. aq. NH_4Cl.

1.4 Inhibition of Enzymes of the Shikimate Pathway.

The shikimate pathway only operates in plants and micro-organisms. The three aromatic amino acids cannot be produced by *de novo* synthesis in animals, but must be obtained from the diet. This makes the shikimate pathway a particularly attractive target for the design of specific enzyme inhibitors. Compounds which fulfil this function would be potential herbicides or antibiotics, of low environmental impact.

Inhibitors are divided into two major classes, irreversible and reversible.[60] Irreversible inhibitors react covalently with an enzyme preventing substrate binding or catalysis. Reversible inhibitors undergo rapid equilibrium binding with the enzyme and are further classified as competitive, uncompetitive or non-competitive, depending on whether they bind to the free enzyme, enzyme-substrate complex or both, respectively.

As an inhibitor target, the most important enzyme in the shikimate pathway is 5-enolpyruvylshikimate-3-phosphate synthase, which catalyses the conversion of shikimate-3-phosphate to 5-EPS-3-P. This enzyme is effectively inhibited by glyphosphate (**106**) (*N*-[phosphonomethyl]glycine),[61] the active ingredient of the broad spectrum herbicide Roundup®.

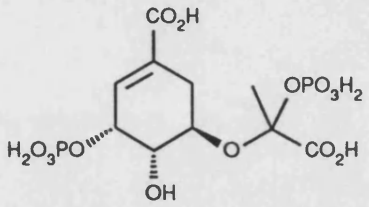


(106)

1.4.1 Synthesis of 5-EPS-3-P Synthase Inhibitors

Bartlett *et al.* have synthesised a number of analogues[62] of the unstable tetrahedral intermediate (**13**), involved in the 5-EPS-3-P synthase reaction. Stable analogues of this high energy intermediate would be expected to benefit from the

extra binding affinity that these species (and transition state structures) experience.[63]



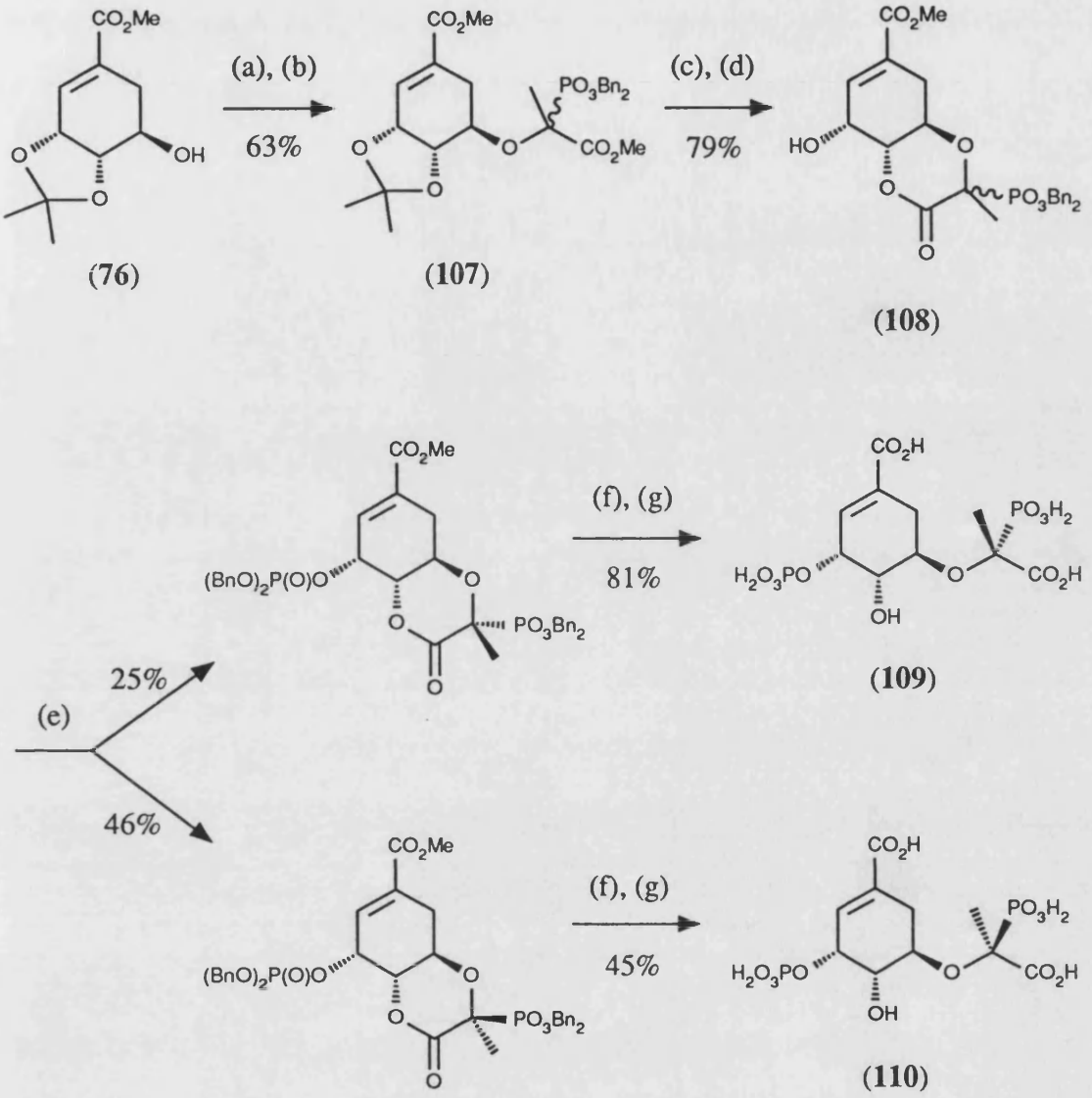
(13)

Two strategies were chosen in order to stabilise the ketal phosphate structure of the tetrahedral intermediate (**13**).

In the first, the phosphate was replaced by a phosphonate moiety. Although the phosphonates produced do not arise from an isosteric replacement of the side chain phosphate, there are examples in which phosphonates are more tightly bound than homophosphonates as replacements for phosphates.[64]

The phosphonates were synthesised from the acetonide of (-)-methyl shikimate (**76**) (**Scheme 1.33**). Rhodium diacetate coupling of (**76**) with methyl (dibenzylphosphono)diazoacetate, followed by methylation, afforded the diastereomeric phosphonates (**107**). Deprotection and cyclisation gave the lactones (**108**), which were phosphorylated prior to chromatographic separation. Deprotection of both diastereomers yielded the phosphonate analogues (**109**) and (**110**), which were purified as their sodium salts.

Both phosphonates (**109**) and (**110**) were shown to be competitive inhibitors of 5-EPS-3-P synthase, with respect to 5-EPS-3-P, with binding constants K_i of 0.015 μM and 1.1 μM respectively. Compound (**109**) is the most potent inhibitor of 5-EPS-3-P synthase yet reported, binding more than a magnitude greater than glyphosphate itself. Since compound (**109**) binds much tighter than (**110**), it was suggested that this infers that the side chain of the natural intermediate (**13**) also has the same absolute configuration.

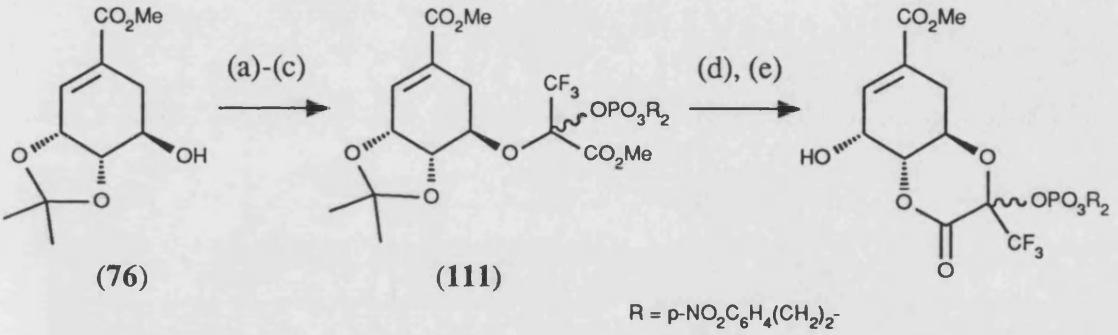


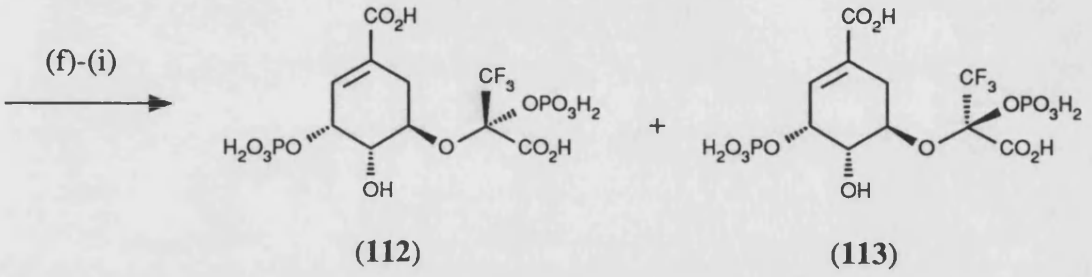
Scheme 1.33 Reagents: (a) $(BnO)_2P(O)C(N_2)CO_2Me$, $Rh_2(OAc)_4$, PhH, reflux; (b) KH, MeI, THF; (c) *p*-TSA, aq. MeCN; (d) *p*-TSA, PhH, reflux; (e) LDA, $[(BnO)_2P]_2O$, THF, -78 to 10°C; (f) TMSBr; (g) aq. NaOH.

The second strategy involved the introduction of electron-withdrawing substituents onto the methyl group of (**13**), in order to destabilise the oxacarbonium ion presumably involved in the decomposition process. The trifluoropyruvate phosphate analogues were synthesised from the acetonide of (-)-methyl shikimate (**76**) (**Scheme 1.34**). Reaction of (**76**) with methyl trifluoropyruvate gave the

hemiketal, which was phosphorylated to afford the diastereomeric phosphates (**111**). Lactone formation, further phosphorylation and deprotection, as before, yielded the trifluoromethyl analogues (**112**) and (**113**).

Again, both compounds were competitive inhibitors of 5-EPS-3-P synthase, with respect to 5-EPS-3P, with binding constants K_i of 0.026 μM and 0.032 μM, respectively. The essentially identical affinity of the two trifluoromethyl analogues, cast doubt on the suggestion that the absolute configuration of the tetrahedral intermediate (**13**), may be determined from the differing affinities of the two phosphonate analogues.

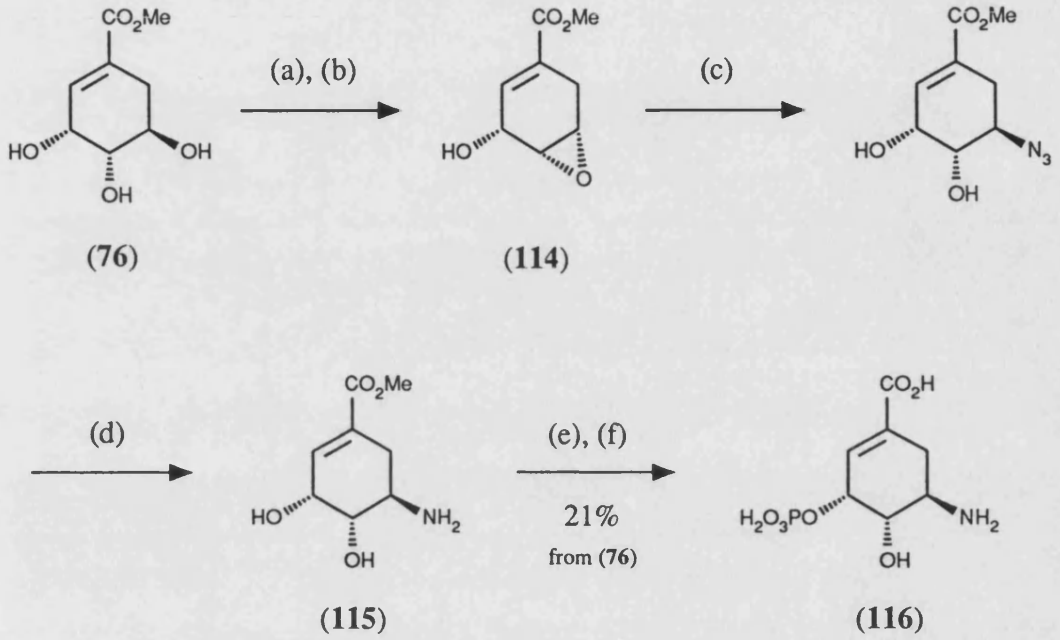




Scheme 1.34 Reagents: (a) $CF_3C(O)CO_2Me$, PCl_3; (b) *p*-$NO_2C_6H_4(CH_2)_2OH$; (c) *m*-CPBA; (d) H_3O^+; (e) K_2CO_3; (f) $[NO_2C_6H_4(CH_2)_2O]_2PNi(Pr)_2$; (g) *m*-CPBA, (h) DBU, BSA; (i) aq. NaOH.

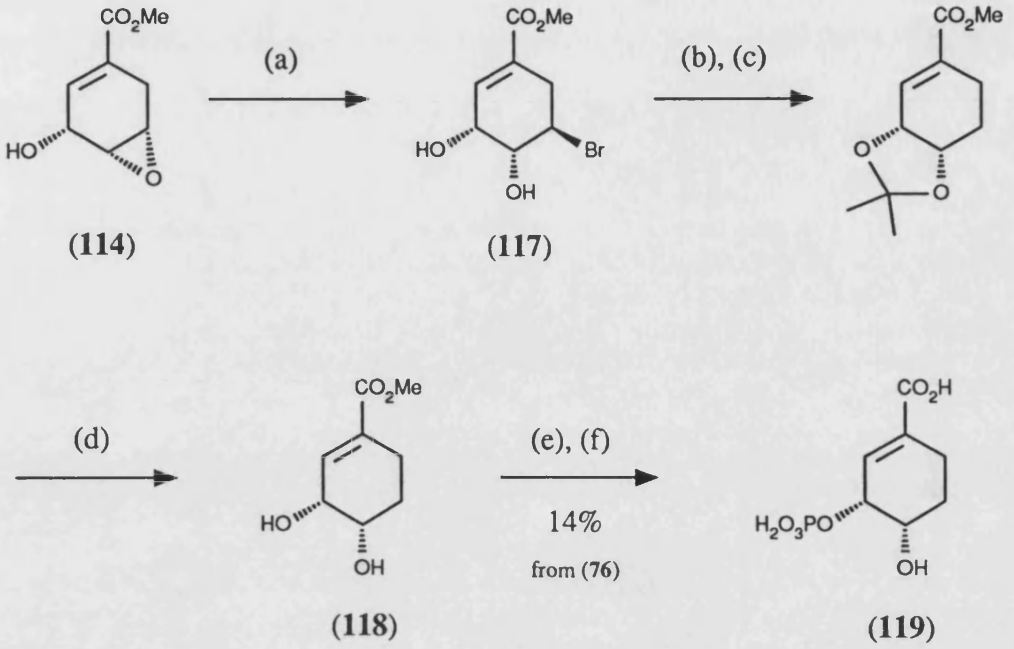
Recently, two new inhibitors of 5-EPS-3-P synthase have been prepared, by Anderson, Knowles *et al.*,[65] to probe the enzyme binding site. The 5-amino analogue

(**116**) was synthesised from (-)-methyl shikimate (**76**) *via* Berchtold's[66] epoxide (**114**) (**Scheme 1.35**). Opening of this epoxide with sodium azide, followed by reduction, furnished the 5-aminoshikimate (**115**). Hydrolysis and enzymatic phosphorylation afforded (**116**), which was a modest competitive inhibitor of 5-EPS-3-P synthase (K_i = 22 μM).



Scheme 1.35 Reagents: (a) Ph_3P, DEAD, THF; (b) 120°C, 0.5 mmHg; (c) NaN_3, NH_4Cl, MeOH, H_2O; (d) H_2, Lindlars cat., EtOH; (e) aq. KOH, THF; (f) ATP, shikimate kinase.

The 5-deoxy analogue (**120**) was also obtained from the epoxide (**114**) (**Scheme 1.36**). Treatment of (**114**) with dilithiotetrabromonickelate provided the bromoshikimate (**117**), which was protected as the acetonide. Tri-*n*-butyltin hydride reduction, followed by deprotection, afforded the diol (**118**). Hydrolysis and enzymatic phosphorylation, as before, gave (**119**), which was a similar competitive inhibitor of 5-EPS-3-P synthase (K_i = 51 μM).



Scheme 1.36 Reagents: (a) Li_2NiBr_4, THF; (b) CH_2=C(OMe)Me, *p*-TSA, CH_2Cl_2; (c) *n*-Bu_3SnH, AIBN, PhH; (d) Dowex 50W-X8 (H^+), MeOH; (e) aq. KOH, THF; (f) ATP, shikimate kinase.

CHAPTER TWO

RESULTS AND DISCUSSION

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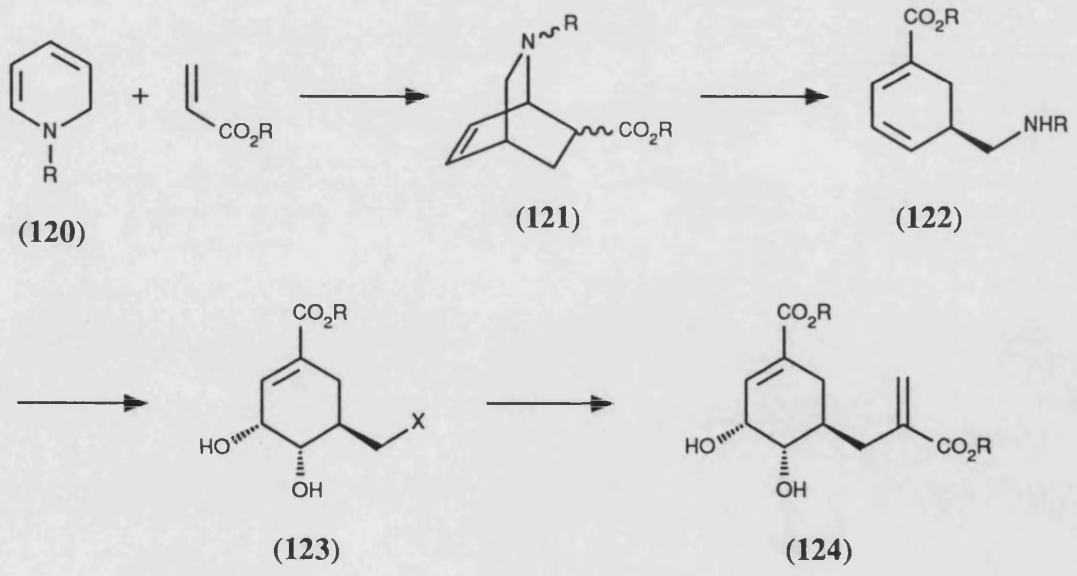
2.1 Aims and Objectives

The primary objective of this project was to synthesise analogues of shikimic acid bearing an additional carbon atom at the 5-position. Such '5-homoshikimates' (**123**) were required for testing as specific enzyme inhibitors.

The synthesis of shikimic acid from furan has been demonstrated both within the Bath laboratory and by other groups (**Section 1.2**). It was envisaged that this approach could be extended by replacing furan with a different heterocycle. Thus, the Diels-Alder reaction of a 1,2-dihydropyridine (**120**), (**Scheme 2.1**), with a suitable acrylate, would be expected to give rise to a mixture of *endo* and *exo* adducts (**121**).



(120) **(121)** **(122)**

(123) **(124)**

Scheme 2.1 Planned Strategy for Synthesis of 5-Homoshikimates

Upon base mediated ring opening these would afford a single (racemic) diene (**122**),

possessing the required carbon framework of the target compounds. Introduction of the *cis*-dihydroxy functionality and modification of the 5-substituent, would yield the desired 5-homoshikimate (**123**).

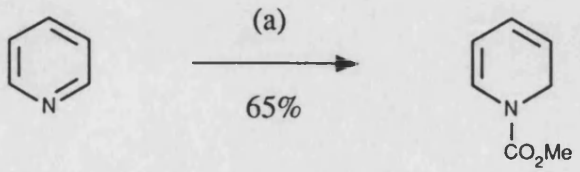
Additionally, further elaboration of the side chain at the 5-position could give access to analogues of later intermediates in the shikimate pathway, such as the carba analogue (**124**), of 5-enolpyruvylshikimic acid.

Dihydropyridines were chosen since the more obvious choice, the oxygen heterocycle 2*H*-pyran, is unknown.[67] In contrast, the chemistry of dihydropyridines has been studied and their usefulness proven in a number of natural product syntheses.[68]

2.2 Synthesis of 5-Homogabaculine

2.2.1 Preparation of the 1,2-Dihydropyridine

This cycloaddition - ring opening strategy was first demonstrated using the 1-methoxycarbonyl-1,2-dihydropyridine (**125**). This compound was prepared from pyridine, according to the procedure of Fowler,[69] (**Scheme 2.2**). Treatment of pyridine with methyl chloroformate in the presence of sodium borohydride, at -78°C, afforded the 1,2-dihydropyridine (**125**) in 65% yield. Even though Fowler has shown[69] that the production of the 1,4-isomer is minimised by performing the reaction at low temperature, a small amount of the 1,4-isomer was detected in the reaction product by TLC analysis.



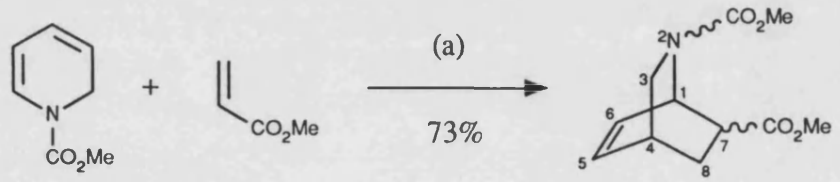
(**125**)

Scheme 2.2 Reagents: (a) MeOCOCl, $NaBH_4$, MeOH, -78°C, 2 h.

N-Alkyloxycarbonyl-1,2-dihydropyridines are considerably more stable than simple *N*-alkyl or unsubstituted examples,[68,69] being less prone to air oxidation. Nevertheless, the carbamate (**125**) discoloured on exposure to air and accordingly it was stored under a nitrogen atmosphere at low temperature. Purification was achieved by chromatography on a short alumina column, eluting with diethyl ether, immediately before use.

2.2.2 Diels-Alder Reaction

The Diels-Alder reaction of 1-methoxycarbonyl-1,2-dihydropyridine (**125**) with methyl acrylate, has been reported by Sliwa,[70] but no experimental details were given. In our hands, the reaction was found to proceed smoothly in refluxing toluene giving the mixed *exo* and *endo* adducts (**126**) as a colourless oil, after column chromatography, in 73% yield (**Scheme 2.3**).



(**125**) (**126**)

Scheme 2.3 Reagents: (a) PhMe, reflux, 3 days.

Analysis of the ^{1}H NMR spectrum indicated that only C-7 substituted products were formed and the *endo*:*exo* ratio was 7:5. The regioselectivity is to be expected from the application of frontier orbital theory[71] to the cycloaddition of a 1-heterosubstituted diene with a dienophile bearing an electron withdrawing group and a consideration of the orbital coefficients in the HOMO of the diene and those in the LUMO of the dienophile. The *endo* preference in cycloadditions with an acrylate ester, results from secondary orbital interactions which provide a small, but significant reduction in the energy of the transition state leading to the *endo* isomer.

The ^1H NMR spectrum of the product is complicated by conformational mobility between the two possible orientations for the *N*-methoxycarbonyl group, for each (*endo* or *exo*) adduct (**Figure 2.1**). Signals due to each conformation are evident in approximately equal proportions, in the spectrum.

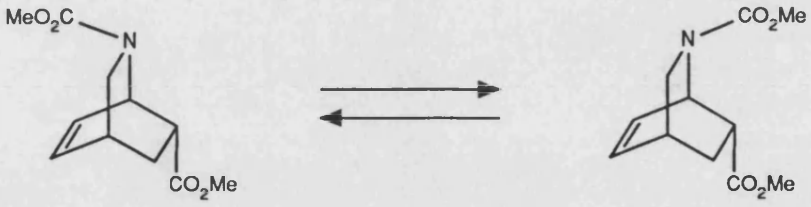


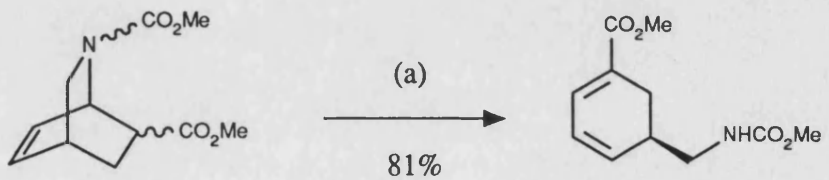
Figure 2.1

The *exo* and *endo* adducts proved inseparable by chromatography, but this was unimportant since ring opening of either adduct leads to the same diene. The mixture was therefore carried forward and used in the next step.

2.2.3 Ring Opening of the Diels-Alder Adducts

The ring opening of 2-azabicylco[2.2.2]oct-5-ene systems may be considered as a reverse Michael addition and, since in the case of the furan adduct 7-oxobicyclo[2.2.1]hept-5-ene, lithium hexamethyldisilazide was found to be the best base to initiate the reaction,[22,23,26] this was also our first choice of reagent.

Treatment of the mixed adducts (**126**) with this reagent (generated by the action of *n*-butyl lithium on 1,1,1,3,3,3-hexamethyldisilazane), produced the diene (**127**), in 81% yield (**Scheme 2.4**).

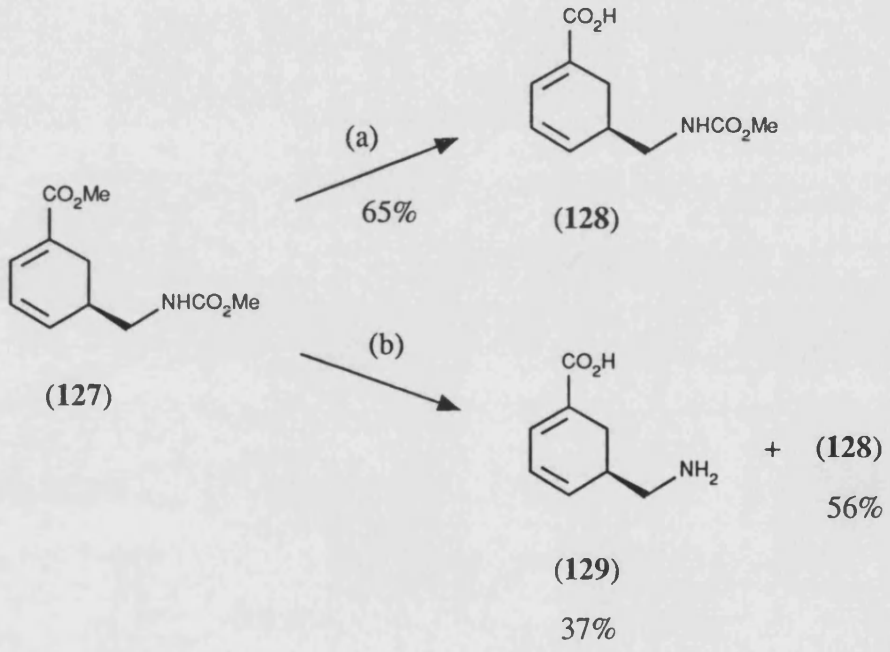


(**126**) (**127**)

Scheme 2.4 Reagents: (a) *n*-BuLi, $(TMS)_2NH$, THF, -78 to 20°C, 20 min.

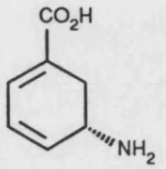
2.2.4 Hydrolysis of the Diene (127)

Alkaline hydrolysis of the diene (**127**) was investigated in order to prepare the amino acid. Treatment of (**127**) with aqueous sodium hydroxide at 20°C produced only the carbamate acid (**128**) (**Scheme 2.5**), but under reflux conditions, hydrolysis of the carbamate function also occurred, giving a mixture of the amino acid (**129**) and the carbamate acid (**128**). Separation was achieved by first acidifying the reaction mixture and extracting the acid carbamate (**128**) into ethyl acetate. The aqueous phase was lyophilised and purified by ion-exchange chromatography to yield the amino acid (**129**).



Scheme 2.5 Reagents: (a) aq. NaOH, THF, 20°C, 16 h; (b) aq. NaOH, THF, reflux, 3 days.

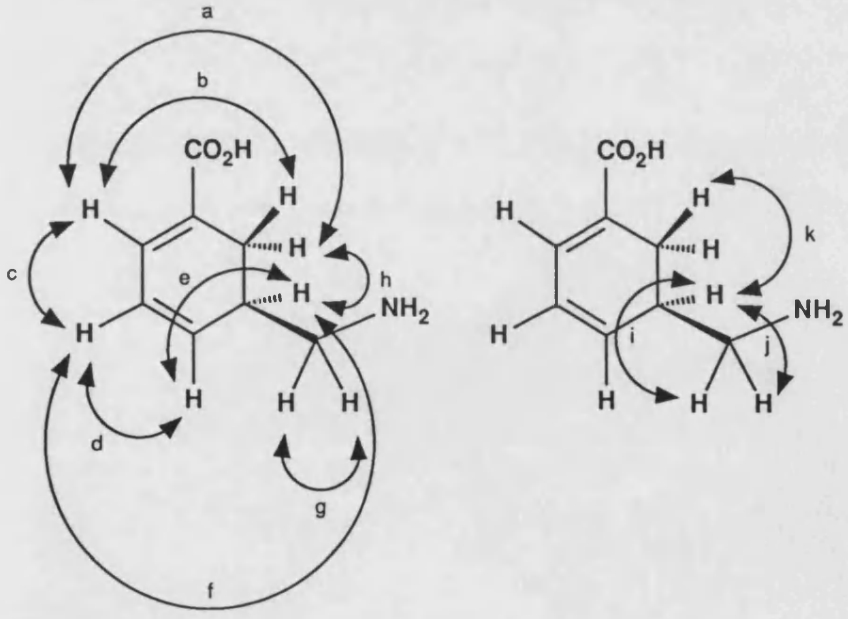
The amino acid (**129**) was named 5-homogabaculine, since it is the 5-homologue of gabaculine. Naturally occurring (-)-gabaculine (**130**), was first isolated from *Streptomyces toyocaensis* by Mishima *et al.*[72] and is a potent inhibitor of γ-aminobutyric acid transferase.



(130)

When this enzyme is inhibited an intracellular build up of γ-aminobutyric acid occurs and this inhibition may be useful in the treatment of Parkinsons disease, epilepsy and Schizophrenia, since these diseases are characterised by a deficiency of this acid. The homologue (**129**), although racemic, was therefore of interest for biological testing.

The ^1H NMR spectrum of the amino acid (**129**) was similar to those of the dienes (**127**),(**128**) and later compounds: several long range and allylic couplings are observed and much work was undertaken, with the aid of 2D COSY experiments, in order to obtain the data which is summarised in **Figure 2.2**.



$J_a = 2.0$ Hz
$J_b = 1.0$ Hz
$J_c = 5.5$ Hz
$J_d = 9.5$ Hz
$J_e = 4.4$ Hz
$J_f = 1.5$ Hz
$J_g = 12.8$ Hz
$J_h = 8.5$ Hz
$J_i = 6.6$ Hz
$J_j = 7.0$ Hz
$J_k = 8.5$ Hz

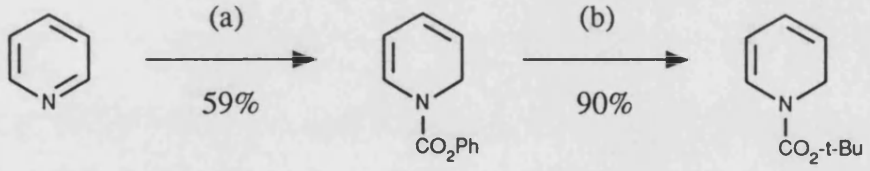
Figure 2.2 Coupling Constants for (**129**) (in D_2O).

2.3 Synthesis of 5-Homoshikimic Acid

An extension of the synthesis required the carbamate to be hydrolysed selectively in the presence of the methyl ester. To this end, a different dihydropyridine starting material was chosen, 1-*tert*-butoxycarbonyl-1,2-dihydropyridine.

2.3.1 Preparation of the 1,2-Dihydropyridine

The required 1,2-dihydropyridine was prepared according to the procedure of Sundberg *et al.*[73] These workers demonstrated that direct acylation of pyridine with di-*tert*-butyl dicarbonate or 2-[[(*tert*-butoxycarbonyl)oxy]imino]-2-phenylacetonitrile, under the reductive conditions which are satisfactory in the case of the acylation with methyl chloroformate, cannot be used. The more obvious reagent, *tert*-butyl chloroformate, is unavailable and so an exchange route was developed (**Scheme 2.6**).



(131) **(132)**

Scheme 2.6 Reagents: (a) PhOCOCl, $NaBH_4$, MeOH, -78°C, 2 h; (b) *t*-BuOK, THF, 20°C, 1 h.

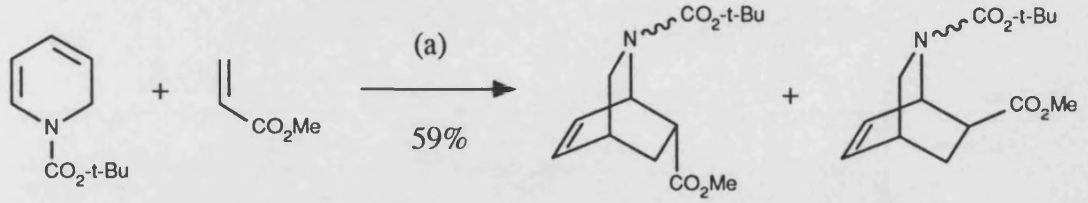
Phenyl chloroformate when reacted with pyridine in the presence of sodium borohydride gave the crystalline dihydropyridine (**131**), in 59% yield. The phenoxy group was then displaced by treatment with 1.1 equivalents of potassium *tert*-butoxide in THF, to give the *tert*-butoxycarbonyl derivative (**132**). The crude yield of this product was 90%, however, the corresponding 1,4-dihydropyridine was

also present as a minor impurity, due to base catalysed isomerisation of (**132**). Sundberg has reported that the use of 2.8 equivalents of potassium *tert*-butoxide gives predominantly the 1,4-isomer, whereas 1.2 equivalents leads to the 1,2-isomer.[73]

2.3.2 Diels-Alder Reaction

The Diels-Alder cycloaddition of 1-*tert*-butoxycarbonyl-1,2-dihydropyridine (**132**) with methyl acrylate was performed under the same conditions as used for the methyl derivative (**Scheme 2.7**). The mixed *exo* and *endo* adducts were obtained in 59% yield, the product ratio being 1:1 (as determined by ^{1}H NMR). No correction was made for the amount of 1,4-dihydropyridine impurity in the starting material and this is reflected in the low yield.

Although inseparable by chromatography, the isomers were separable by crystallisation of the *endo* isomer (**133**) from toluene. On the large scale that this reaction was usually done, the crystallisation was conveniently performed on the crude product (obtained after evaporation of excess methyl acrylate from the reaction mixture). The mother liquor was then chromatographed, to remove any 1,4-dihydropyridine, yielding the *exo* isomer (**134**) as a colourless oil.



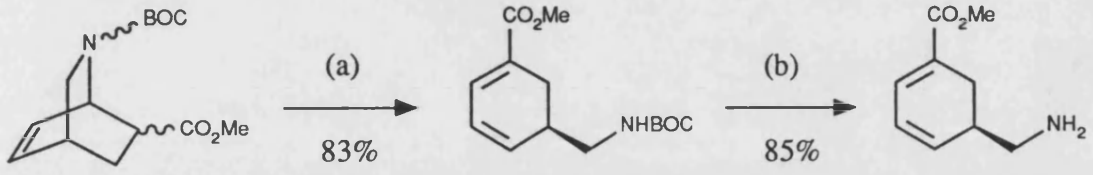
(132) **(133)** **(134)**

Scheme 2.7 Reagents: (a) PhMe, reflux, 2 days.

2.3.3 Ring-Opening and Preparation of the *N,N*-Disulphonyl Derivative

The mixed *exo* and *endo* adducts were ring opened, as before, by treatment with lithium hexamethyldisilazide to afford the diene (**135**) (**Scheme 2.8**). This

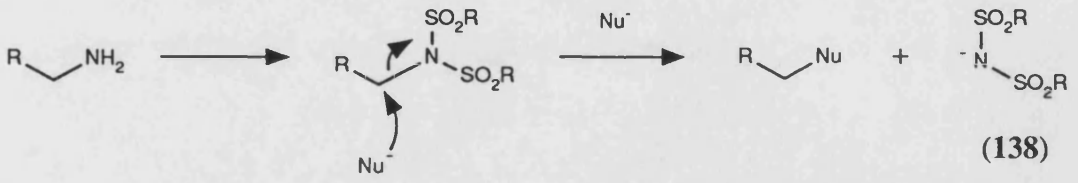
diene has the amino function protected by the *tert*-butoxycarbonyl (BOC) group[74] and thus deprotection was now possible without hydrolysis of the methyl ester. Cleavage of the BOC group using trifluoroacetic acid yielded the amino ester (**136**), in 85% yield.



(**135**) (**136**)

Scheme 2.8 Reagents: (a) *n*-BuLi, $(TMS)_2NH$, THF, -78 to 20°C, 10 min; (b) TFA, 20°C, 5 min;

At this point it was decided to attempt to displace the amino group *via* conversion to a 'good leaving group'. Disulphonimides (**137**) were first shown to be useful intermediates for the deamination of primary aliphatic amines by Baumgarten[75] (**Scheme 2.9**).



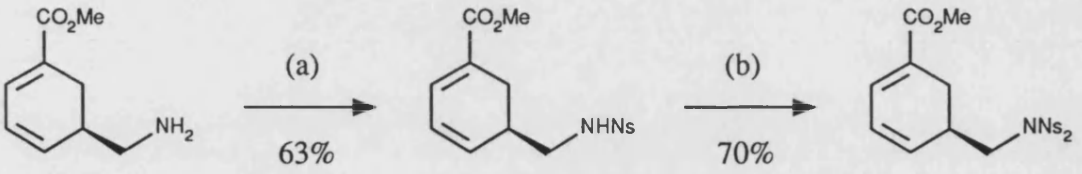
(**137**) (**138**)

Scheme 2.9

The approach is analogous to the substitution of alcohols *via* conversion to the *p*-toluenesulphonyl derivative (tosylate). The disulphonimide ion (**138**) is a relatively weak base and can therefore be displaced by either iodide or bromide ions to yield the corresponding alkyl halides, as well as elimination products. Di-(*p*-nitrobenzene) sulphonimides are less prone to elimination reactions than

di-(*p*-toluene) sulphonimides and thus we decided to use the former.

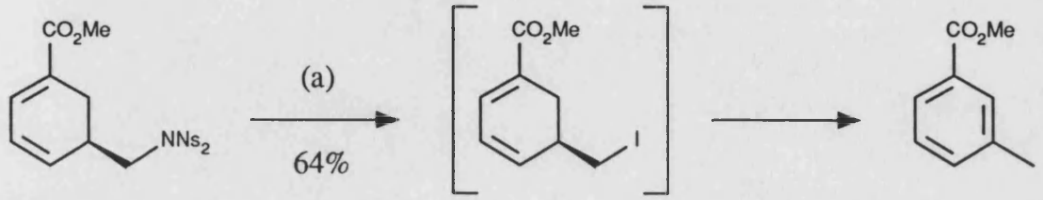
Accordingly the amino ester was treated with triethylamine and *p*-nitrobenzene-sulphonyl chloride (NsCl), thus providing the sulphonamide (**139**) (**Scheme 2.10**). Further sulphonylation using sodium hydride as the base and NsCl afforded the disulphonimide (**140**).



(**136**) (**139**) (**140**)

Scheme 2.10 Reagents: (a) NsCl, Et_3N, THF, 20°C, 40 h; (b) NsCl, NaH, DMF, 20°C, 1.5 h.

The displacement reaction was attempted using Baumgarten's conditions,[75b] namely, heating a solution of the disulphonimide (**140**) in DMF at 110°C with potassium iodide (**Scheme 2.11**). The only product isolated was methyl 3-methylbenzoate (**142**) in 64% yield. Under the same conditions, but with omission of the potassium iodide, no reaction was observed by TLC analysis. Thus, it is probable that displacement did occur to give the iodide (**141**), but that under the reaction conditions this compound then eliminated HI and underwent rearrangement to give the aromatic product (**142**).



(**140**) (**141**) (**142**)

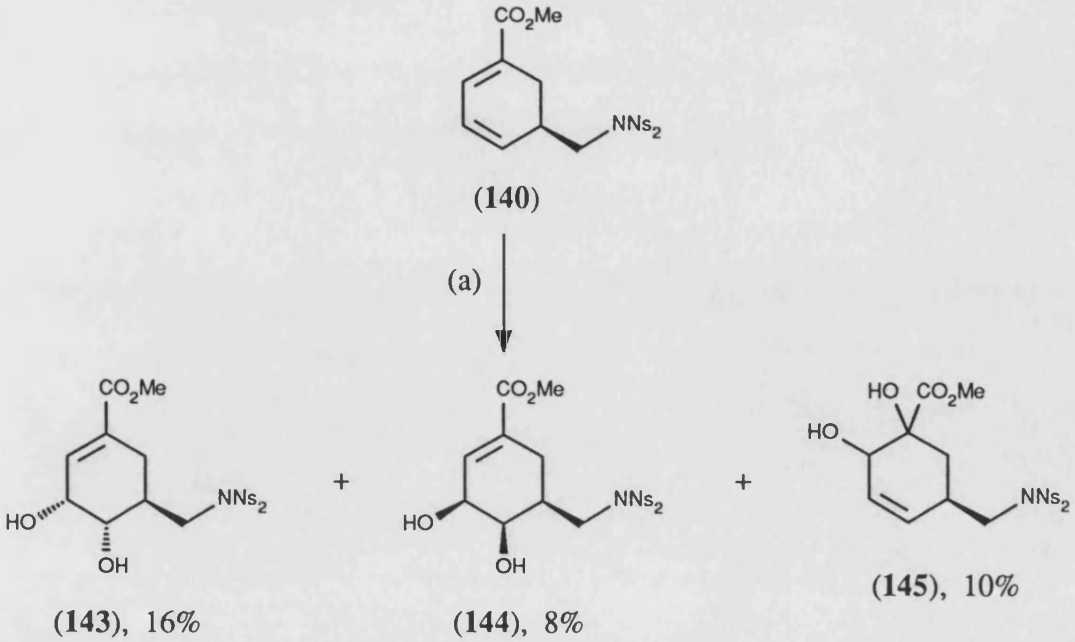
Scheme 2.11 Reagents (a) KI, DMF, 110°C, 16 h.

2.3.4 Osmylation of the Diene Disulphonimide (140)

In view of the results with the diene disulphonimide (**140**), we decided to introduce the 3,4-*cis*-diol functionality before trying the displacement reaction. The driving force of aromatisation would then be absent and substitution products would be more likely.

Osmium tetroxide is the most reliable reagent available for the *cis*-hydroxylation of alkenes.[76] Due to the high toxicity and expense of this reagent, it is more convenient to use it catalytically, in this case a secondary oxidant is employed to oxidatively hydrolyse the intermediate osmium (VI) ester and regenerate the osmium tetroxide, with the reaction being complete on consumption of the secondary oxidant. The use of *N*-methylmorpholine *N*-oxide, rather than hydrogen peroxide, for this purpose was preferred since the latter often gives rise to over oxidised products.[77]

Treatment of the diol disulphonimide (**140**) with *N*-methylmorpholine *N*-oxide and a catalytic amount of osmium tetroxide afforded the three diols (**143**)-(**145**), which were separated by column chromatography (**Scheme 2.12**).



Scheme 2.12 Reagents: (a) OsO_4 (0.05 eq.), NMO, Me_2CO, THF, H_2O, 20°C, 24 h.

The diol with the desired (3α, 4α, 5β) relative stereochemistry, (**143**), was isolated in 16% yield and its structure determined by examination of the ^{1}H NMR spectrum. When this was run in D_6-DMSO, signals due to the diol functionality were immediately obvious in the form of two doublets at δ 4.69 ($J_{4,OH}$ 6.8 Hz) and δ 5.11 ($J_{3,OH}$ 6.2 Hz). Other resonances were more complex, due to unresolved fine coupling interactions, but an assignment was made possible through a 2D COSY experiment. When run in D_6-acetone/D_2O the spectrum was simplified allowing coupling constants to be measured. These are summarised in **Figure 2.3**.

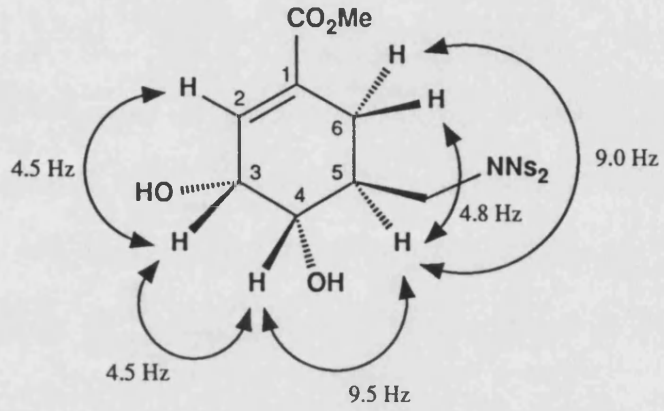


Figure 2.3. Coupling Constants for (**143**) (in D_6-acetone/D_2O)

These figures correlate reasonably well with coupling constants calculated from the Karplus equation.[78] Even though this method needs to be used with caution, especially when electronegative groups are present, it seems to be generally applicable to shikimic acid derivatives and analogues.[79] The geometry of the cyclohexene ring is essentially defined by the double bond, which forces the four carbon atoms (C-6 and C-1 to C-3) to be coplanar. The ring can then adopt either a half-chair or boat conformation, of which the former is generally favoured.

It is important to realise that rapid conformational inversion is possible at room temperature and the observed conformation is a statistical average of all conformations participating in the inversion cycle. However, the nature of the

substituents determines which conformation is favoured energetically and, providing the energy differences are significant, the observed conformation will approximate to the favoured species.

The resonance due to 4-H is especially informative in the spectra of shikimates and in this case a doublet of doublets was evident at δ 3.62 ($J_{4,3}$ 4.5, $J_{4,5}$ 9.5 Hz). The 4,5-coupling is consistent with a half-chair conformation in which the 5-substituent is in an equatorial position and 4-H and 5-H are both axial (**Figure 2.4**). The *cis* arrangement of the 3-hydroxyl and 4-hydroxyl is not in doubt since these groups arose from osmylation, however, this arrangement is also supported by the relatively small 3,4-coupling. The protons in the 6α and 6β positions were also distinguished by their coupling constants with 5-H and these couplings are also in agreement with the conformation shown. The alternative half-chair and both boat conformations cannot support this data.

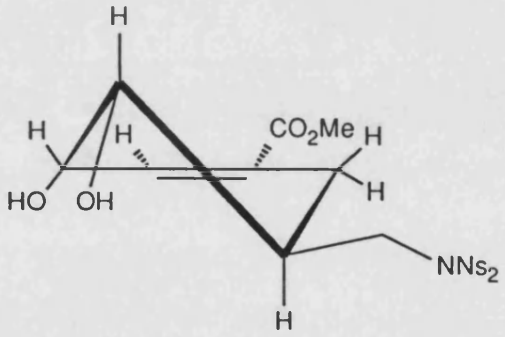


Figure 2.4 Half-Chair Conformation of (**143**).

The other two diols (**144**) and (**145**) were isolated in 8% and 10% yields respectively. Due to their insolubility in $CDCl_3$ or D_6-acetone, NMR spectra could only be obtained for D_6-DMSO solutions and again, assignments were made with the aid of a 2D COSY experiment. The ^1H NMR of (**144**) was similar to that of the diastereomeric diol (**143**), but due to unresolved coupling, the coupling constants could not be measured. The resonance due to 2-H in (**144**) was at higher field (δ

6.53) than that in compound (**143**) (δ 6.70). This difference between the (3β, 4β, 5β) and (3α, 4α, 5β) systems seems a general feature and was followed in the spectra of later compounds.

The relative stereochemistry of the 1,2-diol (**145**) could not be established by ^{1}H NMR. The resonances due to 3-H and 4-H were coincident, as where those due to both 6α-H and 6β-H. Therefore, coupling constants could not be determined and meaningful n.O.e experiments were not possible.

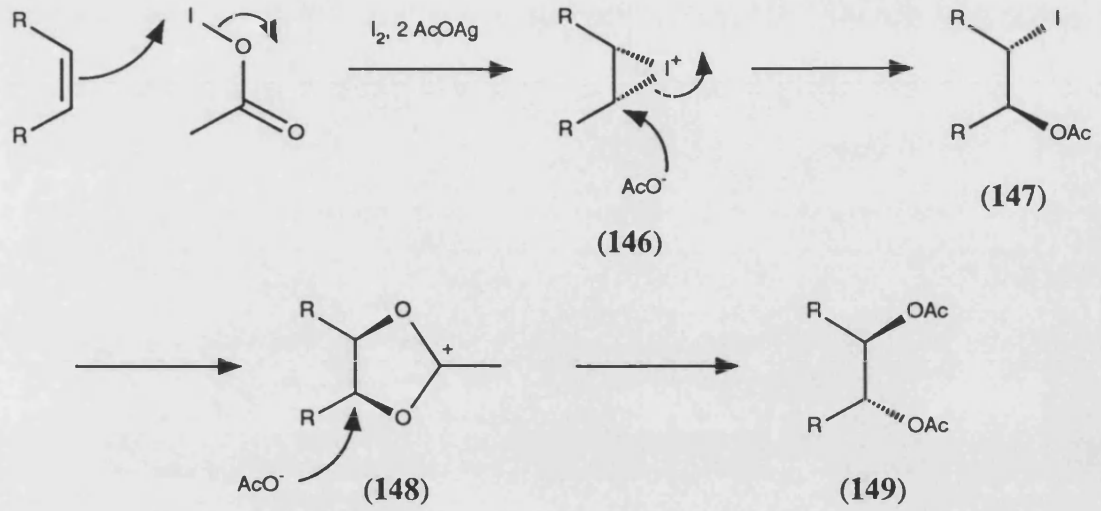
Although all three diols were solids, it was not possible to obtain good quality crystals suitable for an X-ray crystallographic determination, recrystallisation of these disulphonimides consistently produced microcrystalline solids.

The overall yield for the three diols was only 34%, other products were not detected. It is possible that hydroxylation occurred at both double bonds to produce tetra-ols, which were too polar to be eluted from the column during chromatography. Due to the poor yield of the required diol (**143**), an alternative *cis*-hydroxylation method was sought.

2.3.5 'Wet' Prévost Reaction upon the Diene Disulphonimide (140)

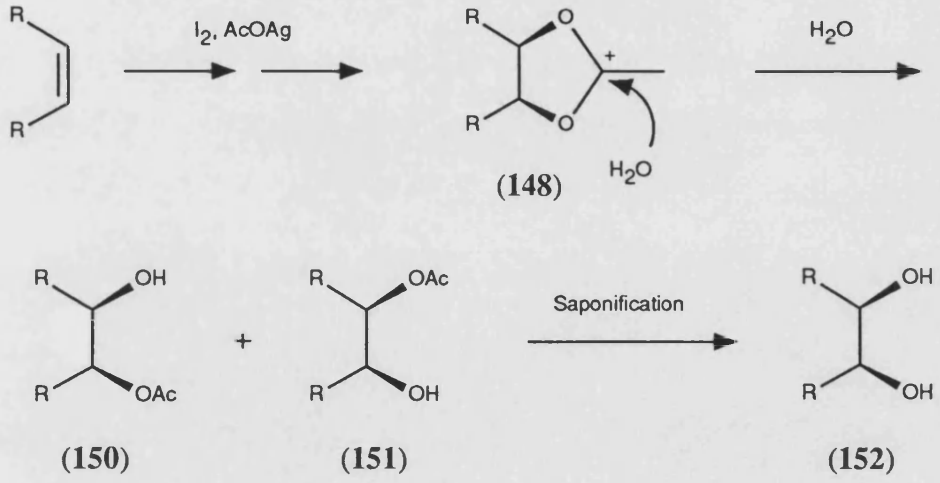
The Prévost reaction involves the reaction between an alkene, iodine and a silver carboxylate (typically the acetate or benzoate) to yield the diester of a *trans* diol (**Scheme 2.13**).[80] The mechanism involves reaction of iodine with two equivalents of silver acetate to afford a complex, AcOAg·AcOI, known as a Simonini complex. Electrophilic attack of the acyl hypoiodite to the least hindered face of the alkene, produces an iodonium species (**146**). Opening of this cation with acetate ion gives an iodo acetate (**147**), which is converted to a cyclic acetoxonium ion (**148**). Finally, further nucleophilic displacement by acetate ion affords a *trans* diacetate (**149**).

A useful modification of the Prévost reaction, the 'wet' Prévost reaction or Woodward hydroxylation,[81] involves the addition of at least one equivalent of water



Scheme 2.13 Mechanism of the Prévost Reaction.

to the reaction mixture (**Scheme 2.14**). The intermediate acetoxonium ion (**148**) undergoes hydrolysis under these conditions, rather than a second nucleophilic displacement, yielding a mixture of *cis* hydroxy acetates (**150**) and (**151**). Subsequent saponification leads to a *cis* diol (**152**) with a stereochemistry corresponding to overall *syn* addition from the more hindered face of the alkene.



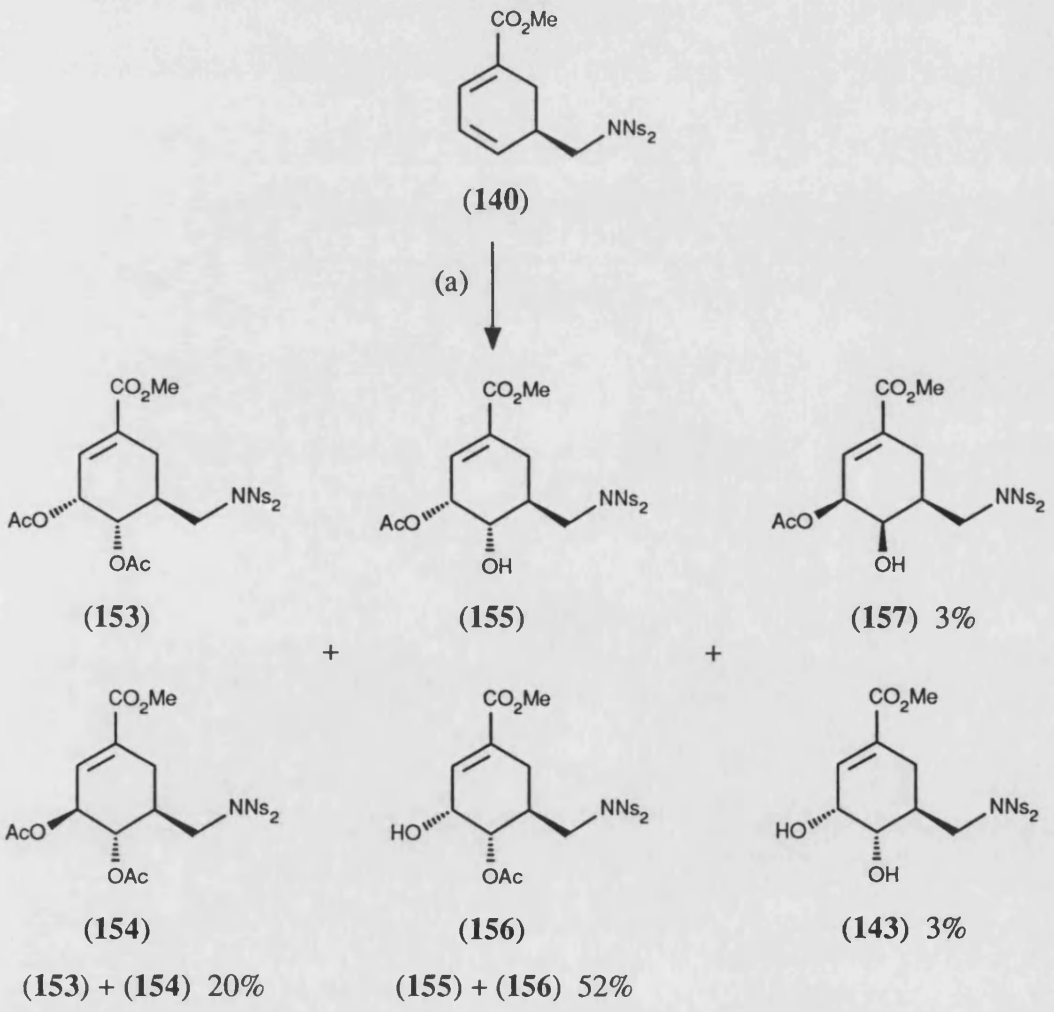
Scheme 2.14 Mechanism of the Woodward Hydroxylation.

Thus, the Woodward hydroxylation is usually complementary to the use of

osmium tetroxide, which gives products resulting from preferential attack from the least hindered face of the alkene.

In practice, the Prévost reaction or Woodward hydroxylation of more highly substituted alkenes, can give rise to a several products. The product ratio is determined by preferential attack of the acyl hypoiodite on the alkene and/or a preferred direction of opening of the acetoxonium ion. The possibility of carbonium ion intermediates can also lead to further complication.

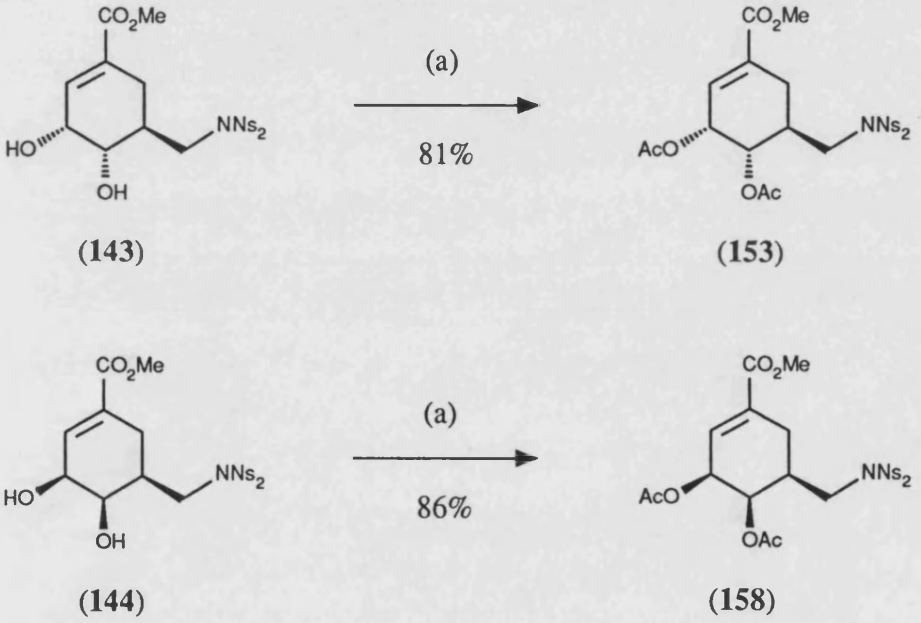
A 'wet' Prévost reaction was performed on the diene disulphonimide (**140**), by treatment with iodine and silver acetate in glacial acetic acid containing an equivalent of water (**Scheme 2.15**).



Scheme 2.15 Reagents: (a) AgOAc, I_2, AcOH, H_2O, reflux, 4 h.

A complex mixture of six products was obtained which was subjected to column chromatography. The epimeric diacetates (**153**) and (**154**) were obtained as an inseparable 1:1 mixture, in 20% yield. The major product was a 1:1 mixture of the hydroxy acetates (**155**) and (**156**), in 52% yield and again these were inseparable by chromatography. Two minor products were also isolated, the hydroxy acetate (**157**) and the previously prepared diol (**143**), both in 3% yield.

The structure of the products was determined by examination of the ^1H NMR spectra. In order to aid identification, the two diacetates (**153**) and (**158**) were also prepared *via* acetylation of the diols (**143**) and (**144**) obtained in the osmylation reaction (**Scheme 2.16**).



(**143**) (**153**)

(**144**) (**158**)

Scheme 2.16 Reagents: (a) Ac_2O, Py, DMAP, 20°C, 24 h.

On examination of the ^1H NMR data for the mixture of diacetates (**153**) and (**154**), obtained from the Prévost reaction, it was obvious that one component was the (3α, 4α, 5β) compound (**153**), identical to the product obtained by acetylation of the diol (**143**). The NMR data for the most useful ring protons (2-H to 4-H), are summarised in **Table 2.1** and support a diaxial arrangement of 4-H and 5-H (**Figure**

2.5). The other diacetate showed a marked difference in the 2,3- and 3,4-coupling constants, but a similar 4,5-coupling constant. The larger 3,4-coupling of 7.1 Hz for compound (**154**) is consistent with (3β, 4α, 5β) stereochemistry, in which the molecule adopts the half-chair conformation shown in **Figure 2.5**.

The ^{1}H NMR data for the major product, a mixture of the hydroxy acetates (**155**) and (**156**), indicated that these have the desired (3α, 4α, 5β) stereochemistry, both showed coupling constants similar to those for compound (**153**) (**Table 2.1**).

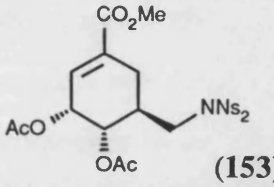
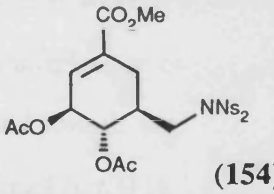
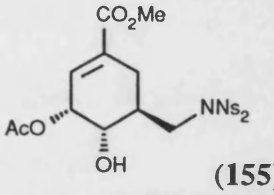
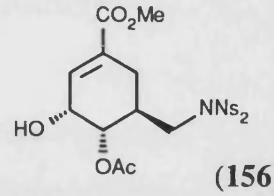
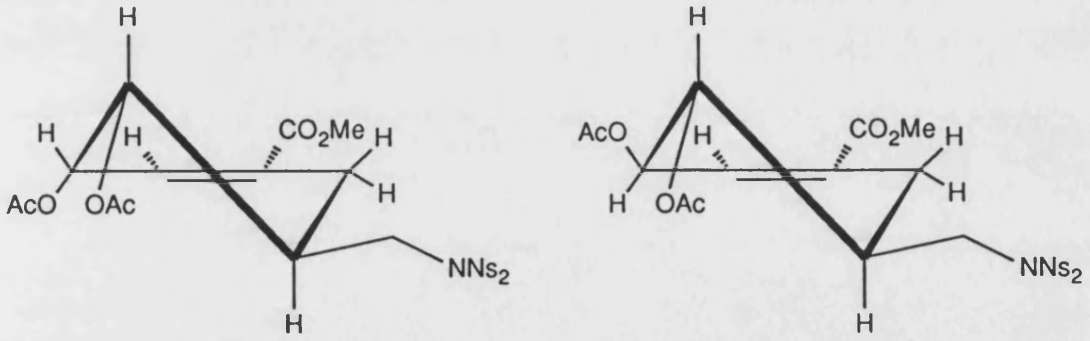
Compound	Chemical shift (δ)			Coupling constant		
	2-H	3-H	4-H	$J_{2,3}$	$J_{3,4}$	$J_{4,5}$
 (**153**)	6.74	5.67	4.97	5.0	4.0	10.1
 (**154**)	6.62	5.56	5.04	2.0	7.1	10.2
 (**155**)	6.79	5.45	4.18	4.6	4.4	11.0
 (**156**)	6.84	4.53	4.87	4.0	4.0	10.0

Table 2.1 ^{1}H NMR Data for the Prévost Reaction Products (**153**) to (**156**).



(153) **(154)**

Figure 2.5 Half-Chair conformations of (**153**) and (**154**).

The final compound to be accounted for was the hydroxy acetate (**157**), a minor product of the reaction. The structure of this compound was assigned by comparison with the ^{1}H NMR data for the (3β, 4β, 5β) diacetate (**158**), obtained by acetylation of the diol (**144**) from the osmylation reaction. The spectra for both these compounds were less informative than for previous compounds. The resonances due to 2-H were broad singlets, whilst those due to 3-H and 4-H appeared as unresolved multiplets or broad singlets. The total width of these multiplets was small, (*ca.* 6 Hz) and thus it was reasoned that the important 2,3-, 3,4- and 4,5-couplings must also small (*ca.* 0 to 3 Hz).

The suggested half-chair conformation for (**158**), that accounts for this data, is shown in **Figure 2.6**. The 5-substituent is equatorial, the 4-substituent axial and consequently the dihedral angles between 2-H, 3-H and 4-H, all give rise to relatively small coupling constants.

An alternative explanation is that since the 4- and 5-substituents cannot both be equatorial, unlike the (3α, 4α, 5β) compounds, the half-chair conformation shown (**Figure 2.6**) is less stable. The energy difference between this and other possible conformations may therefore be slight, leading to time averaged signals and thus broad singlets. However, this would also lead to broadening of the 6-H resonances, which was not observed.

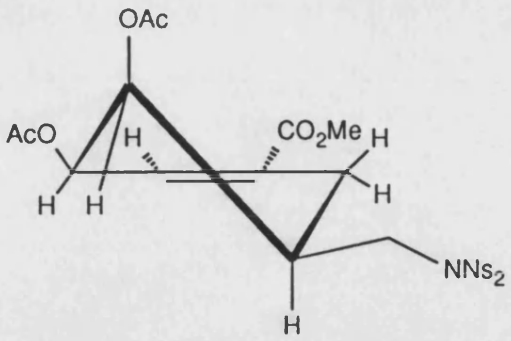
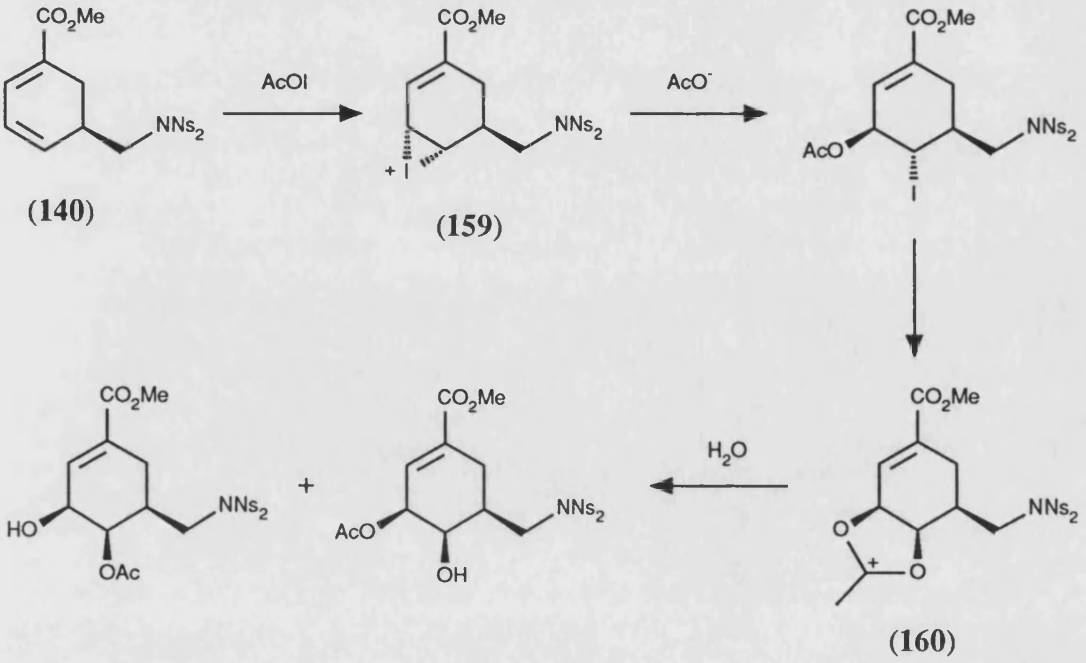


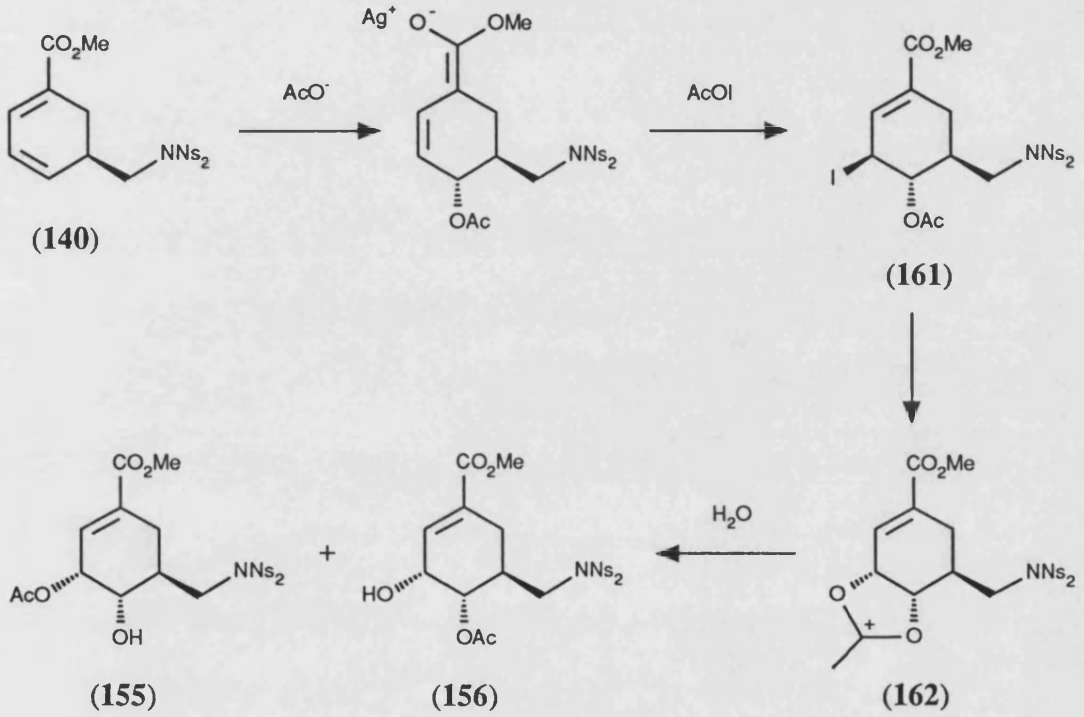
Figure 2.6 Half-Chair conformation of (**158**)

Overall, the reaction was therefore fairly stereoselective, producing products with predominantly the desired (3α, 4α, 5β) stereochemistry. This would not be expected from a consideration of the 'normal' mechanism discussed earlier. The attack of the acyl hypoiodite would be expected from the α-face of the double bond to give the α-iodonium species (**159**) (**Scheme 2.17**). This would then lead to the β-acetoxonium species (**160**) and thus to products having predominantly (3β, 4β, 5β) relative stereochemistry.



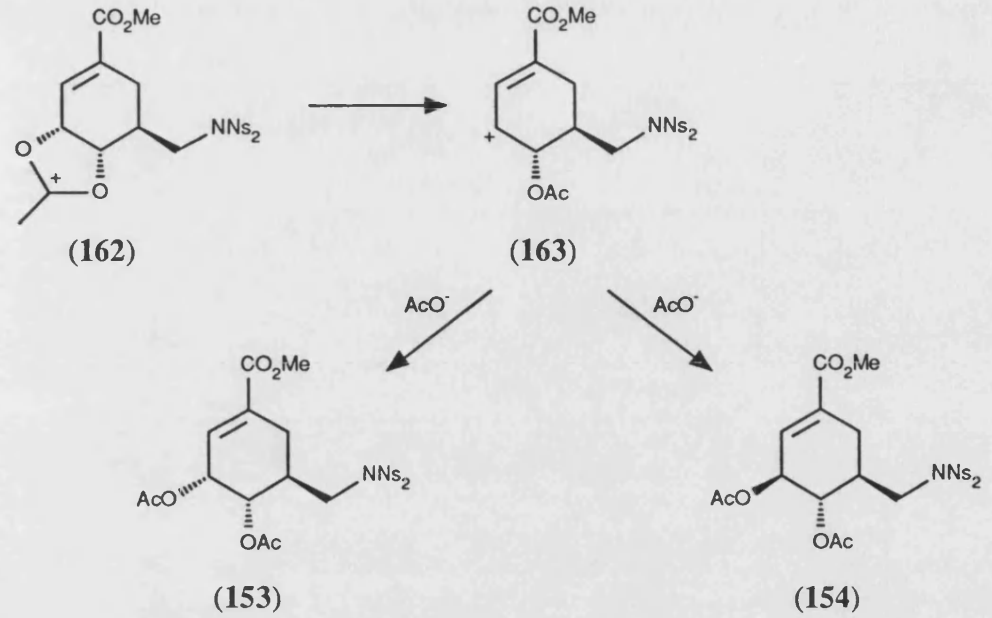
Scheme 2.17

Another possible mechanism has been suggested by Campbell and Sainsbury,[25] to account for this stereoselectivity in Prévost reactions applied to similar dienes. Initial Michael addition by acetate from the α-face at C-4, possibly mediated by the silver cation, followed by electrophilic attack of the acyl hypoiodite, would lead to the iodo acetate (**161**) (**Scheme 2.18**). Cyclisation would give the α-acetoxonium species (**162**) and hence, *via* hydrolysis, the two hydroxy acetates (**155**) and (**156**).



Scheme 2.18

The two diacetates isolated (**153**) and (**154**), would also arise from an α-acetoxonium species. A competing nucleophilic attack of acetate at C-3, rather than hydrolysis, would give the diacetate (**154**). However, since the (3α, 4β, 5β) diacetate, resulting from attack at C-4, is not observed, it is likely that the two diacetates arise through S_N1 attack at C-3, *via* the allylic cation (**163**), rather than S_N2 attack (**Scheme 2.19**).



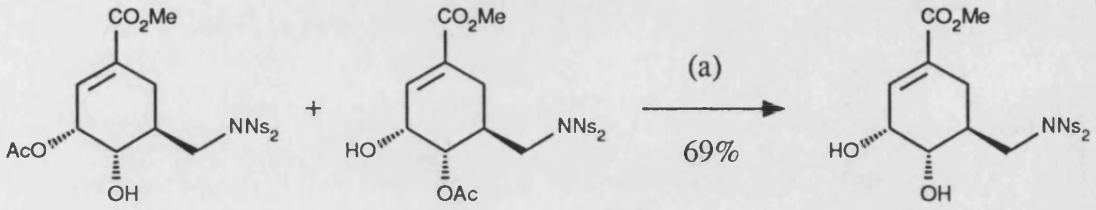
Scheme 2.19

Considering the two minor products isolated, the diol (**143**) would be produced from hydrolysis of either of the hydroxy acetates. The other minor product (**157**), the 3β-acetoxy-4β-hydroxy compound, presumably arises from initial Michael attack of acetate from the β-face of the diene, or from an α-iodonium species. In either case the reaction would proceed *via* a β-acetoxonium intermediate. None of the corresponding 3β-hydroxy-4β-acetoxy compound was isolated, but this fact should not be given too much attention, in view of the small amount of (**157**) isolated.

The low yield of (**157**) from the reaction also testifies to the remarkable selectivity observed for attack to only one face of the double bond.

In summary, it is clear that the major products observed must arise from an α-acetoxonium species (**162**). This may either arise from a conventional Prévost mechanism, but with unexpected preferential formation of the β-iodonium species, or from a different mechanism involving initial Michael attack of acetate ion.

The mixed hydroxy acetates (**155**) and (**156**) were hydrolysed using aqueous ammonia in methanol to afford the diol (**143**) (**Scheme 2.20**). The product was identical, in all respects, to a sample of the diol obtained *via* osmylation, thus confirming the assignments made for the structures of the hydroxy acetates. The overall yield of the diol (**143**) was 36%, for the two steps from the diene disulphonimide (**140**), and in practice this route was much more convenient than using osmium tetroxide.



(**155**) (**156**) (**143**)

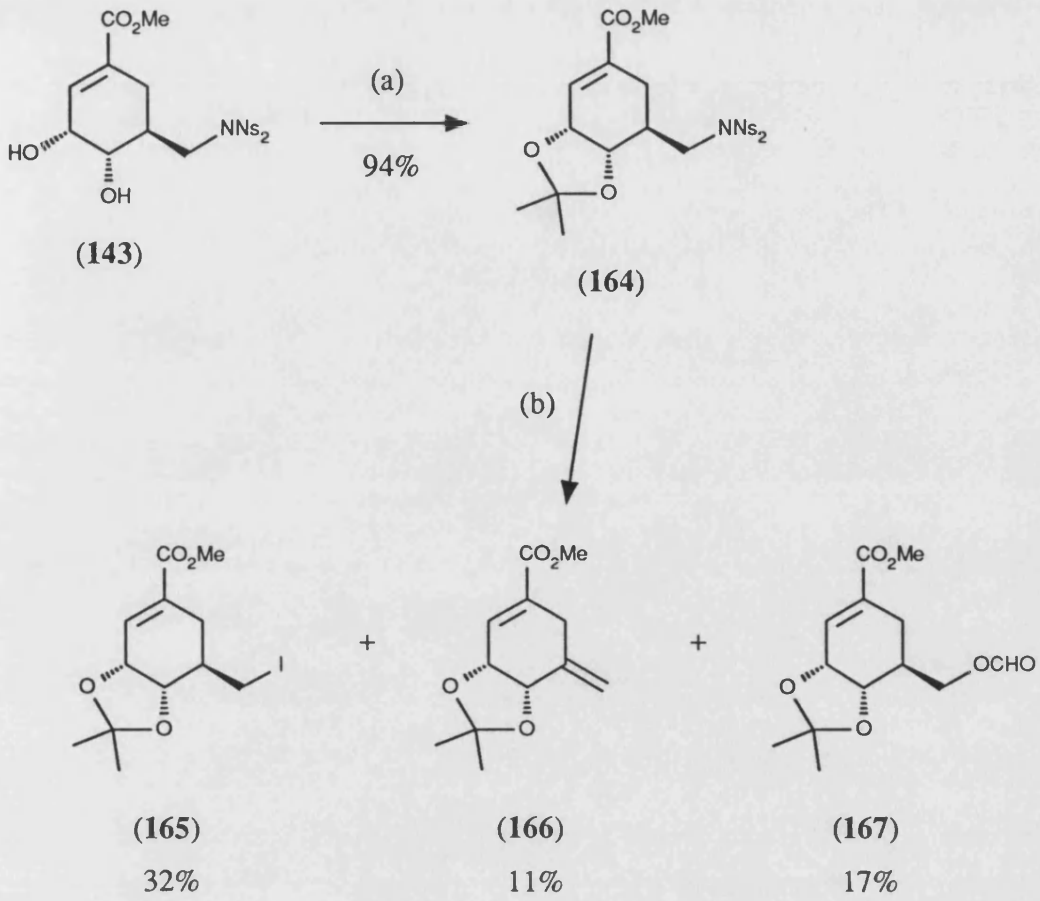
Scheme 2.20 Reagents: (a) aq. NH_3, MeOH, 20°C, 23 h.

2.2.6 Displacement Reactions of the Disulphonimide (164)

With access to sufficient quantities of the diol (**143**) now possible, we could return to the attempted displacement of the disulphonimide group. There were two aims in mind - displacement with iodide to form a useful intermediate for later syntheses and with an oxygen containing nucleophile to give access to the tri-hydroxylated compound.

The two hydroxyl groups of (**143**) were first protected by formation of the acetonide (**164**) (**Scheme 2.21**). Treatment of this compound with potassium iodide in DMF at 130°C, afforded a mixture of three products, which were separated by column chromatography.

The desired iodide (**165**) was isolated as a crystalline solid in 32% yield and the diene (**166**), arising from elimination of HI from (**165**), as a colourless oil, in 11% yield. Recrystallisation of the iodide (**165**) from light petrol (b.p. 40°C), gave



Scheme 2.21 Reagents: (a) $Me_2C(OMe)_2$, Me_2CO, *p*-TSA, 20°C, 22 h; (b) KI, DMF, 130°C, 21 h.

crystals of suitable quality for a successful X-ray crystallographic determination (**Figure 2.7**, full details are included in **Appendix One**). The results confirmed the relative stereochemistry assigned to these compounds on the basis of ^1H NMR data. The conformation of compound (**165**) in the crystal structure was the same as would be expected in solution from ^1H NMR data. The molecule exists in the now familiar half-chair conformation, in which the 5-iodomethyl substituent is equatorial and thus both 4-H and 5-H are axial, leading to a relatively large (9.2 Hz) 4,5-coupling constant (**Figure 2.8**).

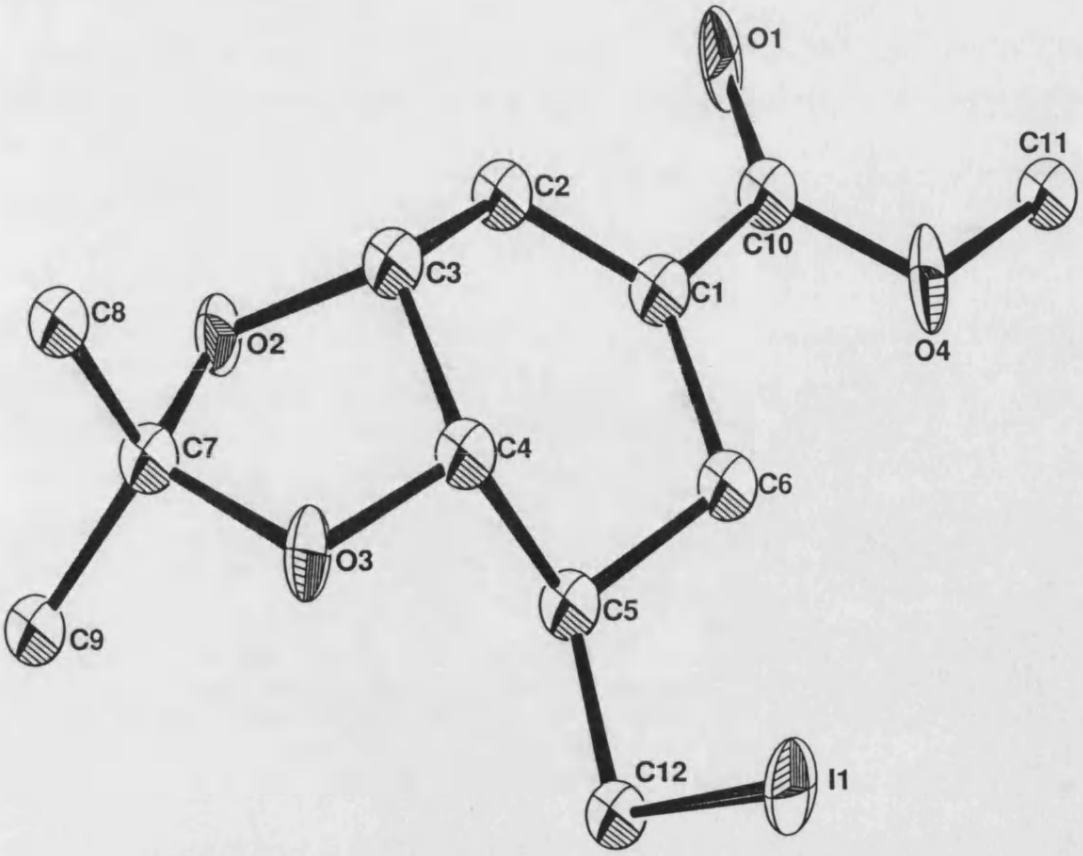


Figure 2.7 ORTEP diagram of (**165**)

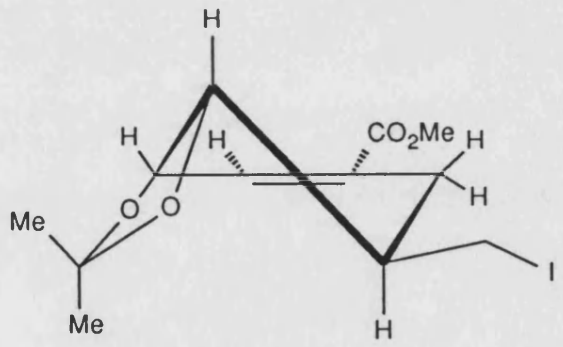


Figure 2.8 Half-Chair conformation of (**165**)

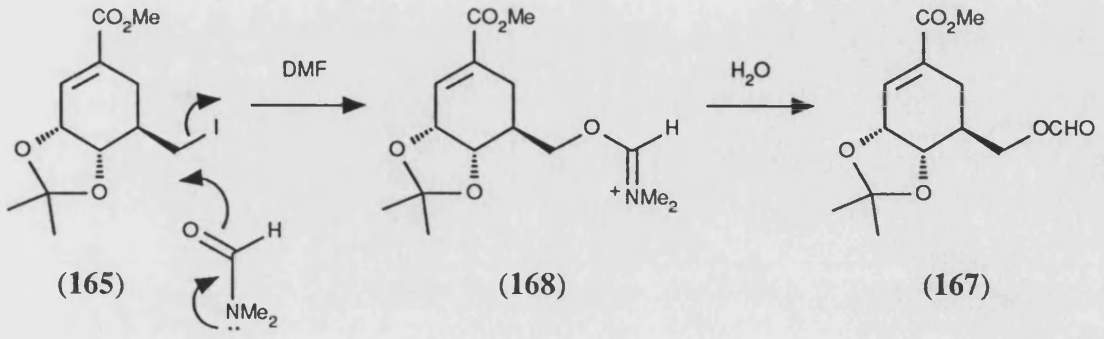
Full ^{1}H NMR data for this compound were obtained by a series of homonuclear decoupling experiments. The resonances due to the proton in the 6β position appeared originally as a sixteen line signal (dddd). When irradiation was carried out at the position corresponding to 2-H, the resonance simplified to eight lines (ddd). Irradiation at the 3-H position produced a similar effect. In this way the

fine allylic (2, 6β) and homo-allylic (3, 6β) couplings were measured. The resonance due to 6α-H was less well resolved and appeared as a broadened doublet of doublets. Irradiation at either the 2-H or 3-H positions produced a marked sharpening of the 6α-H resonance, indicating similar fine couplings to this proton, which are not quite resolved at 270 MHz. Tabulated data is included in the experimental chapter, **Section 3.2**.

The final product obtained was the *O*-formyl derivative (**167**), isolated as a colourless oil, in 17% yield. The *O*-formyl group was evident from the NMR data, giving rise to a low field doublet (*J* 0.5 Hz) at δ 8.10 in the ^{1}H spectrum and a distinct carbonyl resonance at δ 160.8 in the ^{13}C spectrum. Homonuclear decoupling experiments were also carried out on compound (**167**) in order to determine the fine allylic and homo-allylic couplings (these are tabulated in **Section 3.2**).

This compound was of great interest since we had unwittingly introduced a protected hydroxyl group at the 1′-position and thus achieved the synthesis of the protected form of one of our major targets.

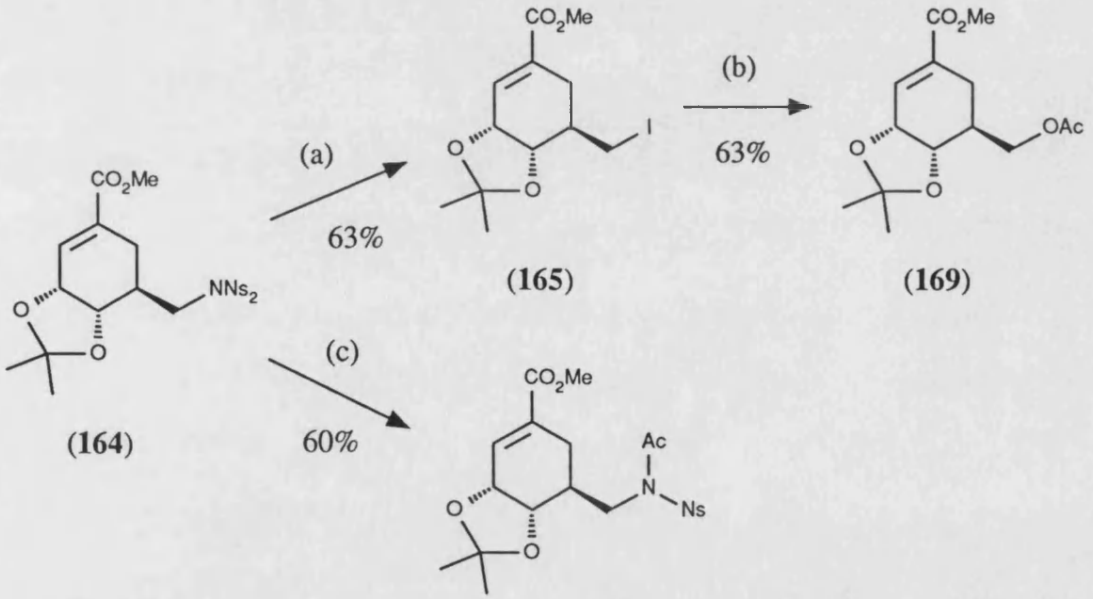
A mechanism for the formation of this product is illustrated in **Scheme 2.22**. Nucleophilic attack of the solvent DMF, on the iodide (**165**), would give the immonium species (**168**). The disulphonimide starting material (**164**) is less likely to undergo this reaction. Hydrolysis of (**168**), during the aqueous workup, would give the *O*-formyl product (**167**).



Scheme 2.22

A similar preparation of formate esters from disulphonimides has been described by Baumgarten.[82] Various simple *N*-alkyl-*N,N*-disulphonimides, when heated with aqueous HI in DMF, gave rise to a mixture of the corresponding alcohols, formate esters and elimination products.

A superior route to the iodide (**165**), from the disulphonimide (**164**), was developed using potassium iodide with 18-crown-6 in toluene (**Scheme 2.23**). Substitution reactions using crown ethers and potassium iodide, have been demonstrated for alkyl tosylates and mesylates,[83] the crown ether forms a complex with the potassium cation, thus solubilising the potassium iodide in the non-polar solvent. The iodide anion is not solvated and consequently much more nucleophilic than in polar solvents. The iodide (**165**) was obtained in 63% yield, and in contrast to the previous method, no elimination product was observed.

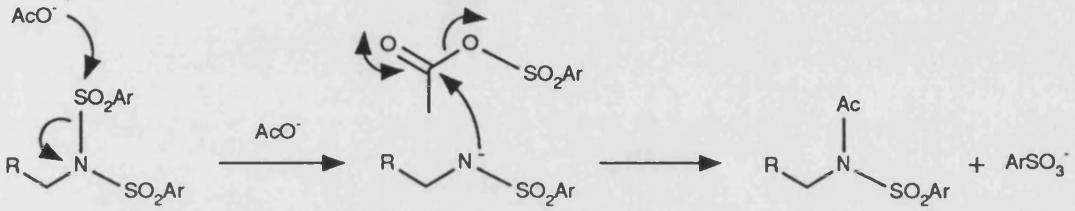


Scheme 2.23 Reagents: (a) KI, 18-crown-6, PhMe, reflux, 7 days; (b) NaOAc, DMF, 110°C, 2.5 h; (c) KOAc, 18-crown-6, PhMe, reflux, 18 h.

With a suitable route to the iodide (**165**) established, the other concern was

to investigate substitution with oxygen nucleophiles. Treatment of the iodide (**165**) with sodium acetate in DMF, yielded the acetate (**169**), in 60% yield (**Scheme 2.23**). The reaction was complete after only 2.5 h, no elimination products were observed.

It was hoped to introduce the acetate group directly *via* treatment of the disulphonimide (**164**) with the 18-crown-6/potassium acetate complex (**Scheme 2.23**). However, in this case the only product produced was the *N*-acetyl derivative (**170**). A mechanism for this transformation in shown in **Scheme 2.24**. Initial nucleophilic attack at sulphur would result in sulphur-nitrogen bond cleavage and formation of the sulphonamide anion (**171**). This species is then acetylated to give the product (**170**) and *p*-nitrobenzenesulphonate anion.



(171)

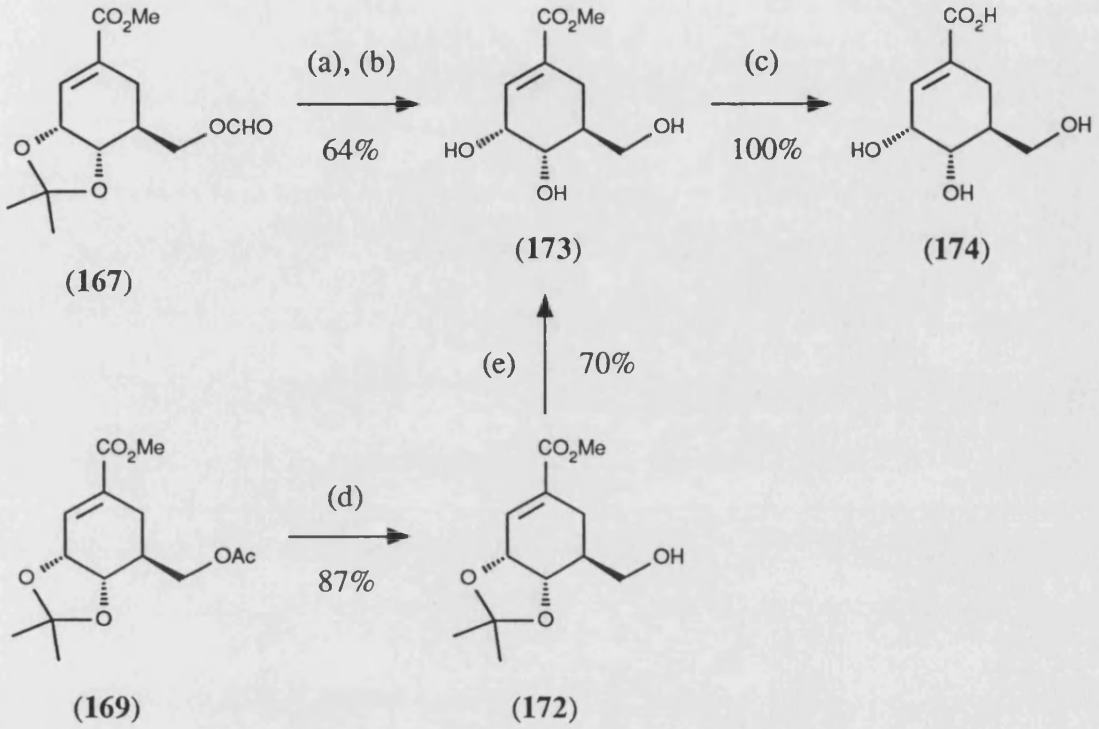
Scheme 2.24

This sulphur-nitrogen rather than carbon-nitrogen bond cleavage in disulphonimides, has also been observed by Baumgarten,[75c,82] and the direct conversion of disulphonimides to alcohols or esters is not possible. This conversion has been described by Andersen,[84] who treated a disulphonimide with a mixture of potassium iodide and potassium acetate in HMPA, to afford the corresponding acetates. Potassium iodide was essential for reaction and the acetates are therefore formed by substitution of the intermediate iodide.

This last reaction was not attempted with our disulphonimide (**164**), however, our sequence did give access to the acetate (**169**) in reasonable yields, by utilising the two step procedure *via* the iodide (**165**), thus the approach is basically the same.

2.3.7 Deprotection

The final stage in the synthesis of our target compound, 5-homoshikimic acid (**174**), was the removal of all the protecting groups from either the formate (**167**), or acetate (**169**) (**Scheme 2.25**). Both of these compounds were used to prepare 5-homoshikimic acid, although the route *via* the acetate (**169**) was preferred, since this compound could be obtained in better yield than the formate (**167**).



Scheme 2.25 Reagents: (a) Amberlyst-15, MeOH, 20°C, 17 h; (b) 50% aq. AcOH, THF, 60°C, 3.5 h; (c) NaOH, H_2O, 20°C, 5.5 h; (d) aq. NH_3, MeOH, 48 h; (e) 50% aq. AcOH, THF, 60°C, 17 h;

Methanolysis of the *O*-formyl group of compound (**167**), using an acidic ion-exchange resin and methanol, also effected incomplete cleavage of the isopropylidene group, affording a mixture of the acetonide alcohol (**172**) and the triol (**173**). The two protecting groups were more conveniently removed in a 'one-pot' reaction, before purification, using aqueous glacial acetic acid to complete

removal of the isopropylidene group. In this way the triol (**173**) was obtained in 64% yield.

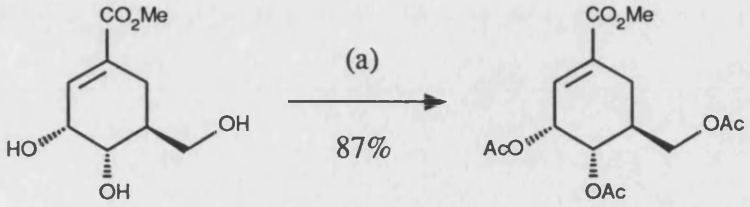
Alternatively, Methanolysis of the acetate (**169**) was performed using aqueous ammonia in methanol to give the acetonide alcohol (**172**), in 87% yield. Removal of the isopropylidene group, again using aqueous glacial acetic acid, afforded the triol (**173**), in 73% yield.

This triol, methyl 5-homoshikimate, (**173**) was saponified using aqueous sodium hydroxide to yield 5-homoshikimic acid (**174**). An acidic ion-exchange resin was used to work-up this saponification and removal of the resin by filtration, followed by lyophilisation, gave excellent yields of the pure acid (**174**). This completed the synthesis of the 5-homologue of shikimic acid, samples of which were submitted for biological testing.

The resonances in the ^{1}H NMR spectrum of 5-homoshikimic acid were assigned with the aid of homonuclear decoupling experiments (tabulated in **Section 3.2**). The signals due to 4-H and one of the 1′-H overlapped in the original spectrum, however, irradiation at the position corresponding to 3-H, simplified the 4-H resonance and allowed the couplings in the 1′-H resonance to be measured.

In order to complete the ^{1}H NMR study of these 5-homoshikimate systems, the triacetyl derivative of methyl 5-homoshikimate was prepared (**Scheme 2.26**). Acetylation of methyl 5-homoshikimate (**173**) with acetic anhydride in pyridine, afforded the triacetate (**175**), in 87% yield.

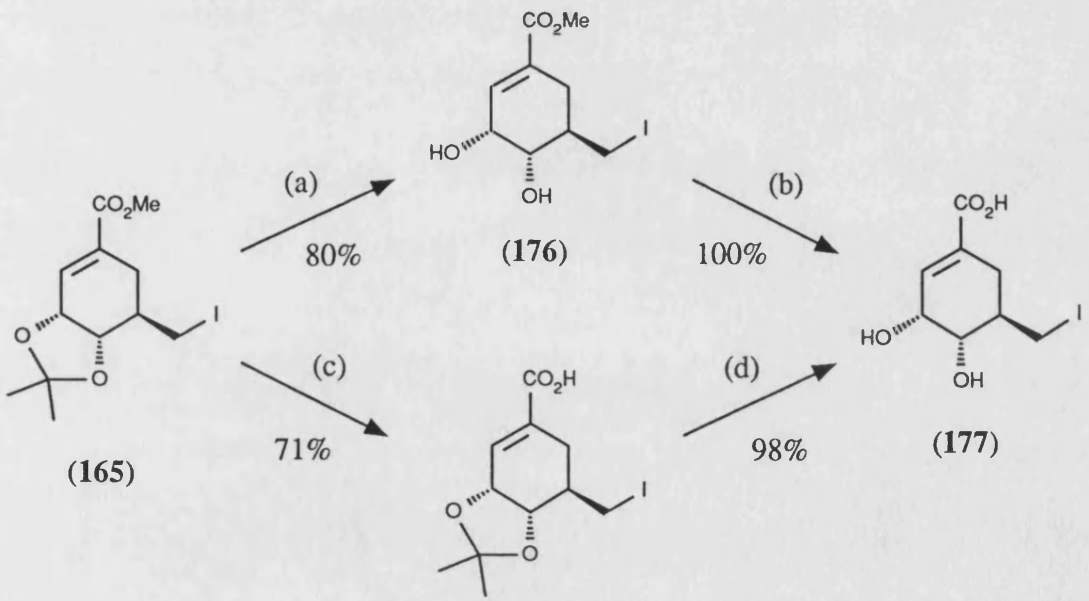
All resonances in the ^{1}H NMR spectrum of this compound were distinct and full assignments could be made. The compound exhibited typical fine allylic coupling ($J_{2,6\alpha}$ 1.3, $J_{2,6\beta}$ 2.4 Hz) and homo-allylic coupling ($J_{3,6\beta}$ 1.2 Hz). The resonance due to 4-H is observed as a doublet of doublets ($J_{4,5}$ 10.8, $J_{4,3}$ 3.9 Hz). The 4,5-coupling is consistent with the expected diaxial arrangement for 4-H and 5-H, although slightly larger than observed in the spectra of previous compounds, a feature due to the presence of the more electronegative acetyl groups.



(173) **(175)**

Scheme 2.26 Reagents: (a) Ac_2O, Py, DMAP, 20°C, 15 h.

Another compound thought suitable for biological testing, was obtained by deprotection of the iodide (**165**) (**Scheme 2.27**). Hydrolysis of the acetonide using aqueous acetic acid yielded the diol (**176**) which was subsequently saponified with aqueous sodium hydroxide to afford the acid (**177**). The order of reactions was also reversed - treatment of (**165**) with aqueous sodium hydroxide giving the acid (**178**) which was converted to (**177**). The iodo substituent was unaffected throughout these reactions although the final product (**177**), did decompose on prolonged storage.



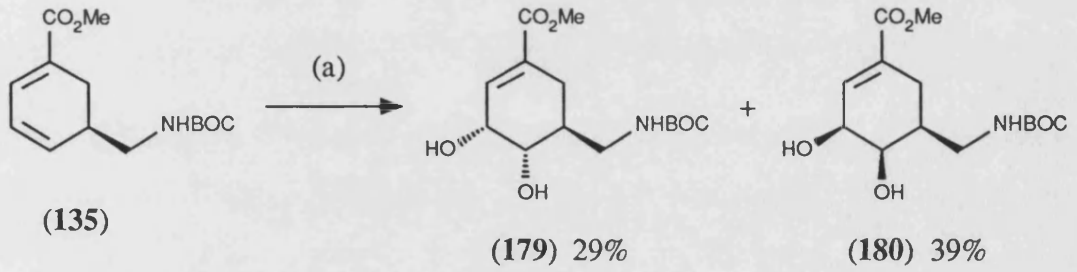
Scheme 2.27 Reagents: (a) 50% aq. AcOH, THF, 60°C, 39 h; (b) NaOH, H_2O, 20°C, 24 h; (c) NaOH, H_2O, 20°C, 18 h; (d) 50% aq. AcOH, THF, 60°C, 24 h.

2.4 Improved Synthesis of 5-Homoshikimic Acid

Although the preceding work had led to a synthesis of our major target, 5-homoshikimic acid (**174**), the approach was somewhat involved, requiring a total of thirteen to fifteen steps (depending on the route used), from pyridine. Considerable interest in the results of biological testing of (**174**), led us to investigate a more concise route to this compound. Starting from the previously prepared diene (**135**), available in four steps from pyridine, the three key stages of hydroxylation, deamination and deprotection were addressed as follows.

2.4.1 Osmylation of the Diene (135)

Treatment of the diene (**135**) with *N*-methylmorpholine *N*-oxide and a catalytic amount of osmium tetroxide afforded a mixture of the diastereomeric diols (**179**) and (**180**) (**Scheme 2.28**). The ratio of (**179**):(**180**) was determined to be 5:4 by examination of the ^{1}H NMR spectrum of the crude product.



(**135**)

(**179**) 29%

(**180**) 39%

Scheme 2.28 Reagents: (a) OsO_4 (cat.), NMO, Me_2CO, H_2O, 20°C, 48 h.

This reaction had been tried earlier in the project, but the two diols proved inseparable by TLC or column chromatography and thus the alternative strategy was developed. On returning to this reaction, it was found that the two products could be separated by trituration with diethyl ether. The diol (**179**) was virtually insoluble in ether and was obtained as a colourless solid which could be recrystallised from ethyl acetate. In contrast, the diastereomeric diol (**180**) was readily soluble in ether and

was obtained as a colourless oil. In this way, separation was possible and the diastereomeric diols could be obtained pure, relatively easily.

The relative stereochemistry of the two products was determined by consideration of the ^{1}H NMR spectra. When the spectrum of (**179**) in D_6-DMSO was examined the resonance due to 4-H was observed as a multiplet at δ 3.39, and the associated coupling constants could not be determined. Using D_6-DMSO as the solvent results in the appearance of distinct doublets for the signals due to the hydroxyl protons and thus it was the coupling between the 4-OH and 4-H that was complicating the 4-H signal. Homonuclear decoupling experiments were used to determine these couplings (tabulated in **Section 3.2**). Irradiation at the resonance due to 3-H, simplified the 4-H resonance to a doublet of doublets ($J_{4,OH}$ 5.5, $J_{4,5}$ 8.5 Hz). Similarly, irradiation at the resonance due to the 4-hydroxyl, produced another doublet of doublets ($J_{4,3}$ 3.5, $J_{4,5}$ 8.5 Hz). The relatively large 4,5-coupling is consistent with a *trans* diaxial arrangement for these two protons and thus (3α, 4α, 5β) stereochemistry.

Recrystallisation of (**179**) from ethyl acetate provided crystals suitable for an X-ray crystallographic determination, which duly confirmed our assignment of this structure (**Figure 2.10**, full details are included in **Appendix Two**). The molecule adopts the same half-chair conformation (**Figure 2.11**) in the crystal structure, as predicted in solution from a consideration of the ^{1}H NMR data.

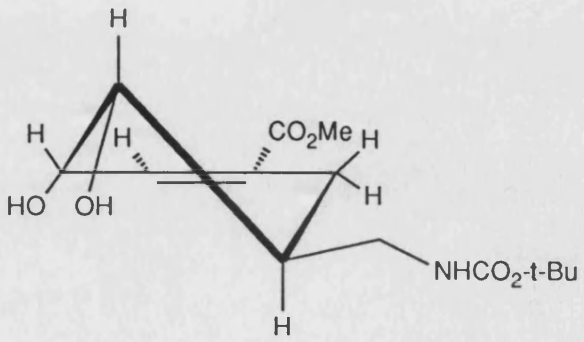


Figure 2.11 Half-Chair Conformation of (**179**)

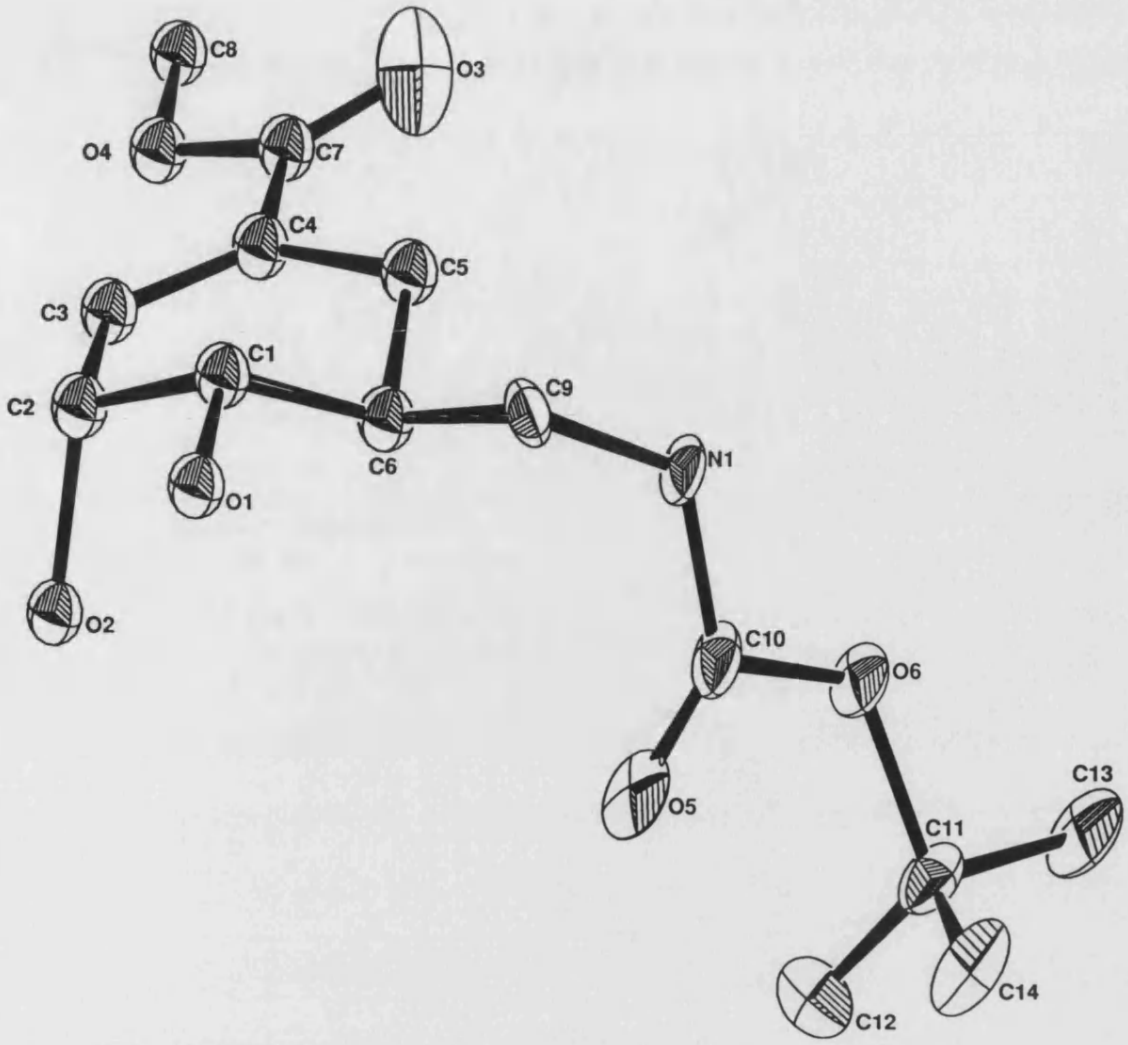


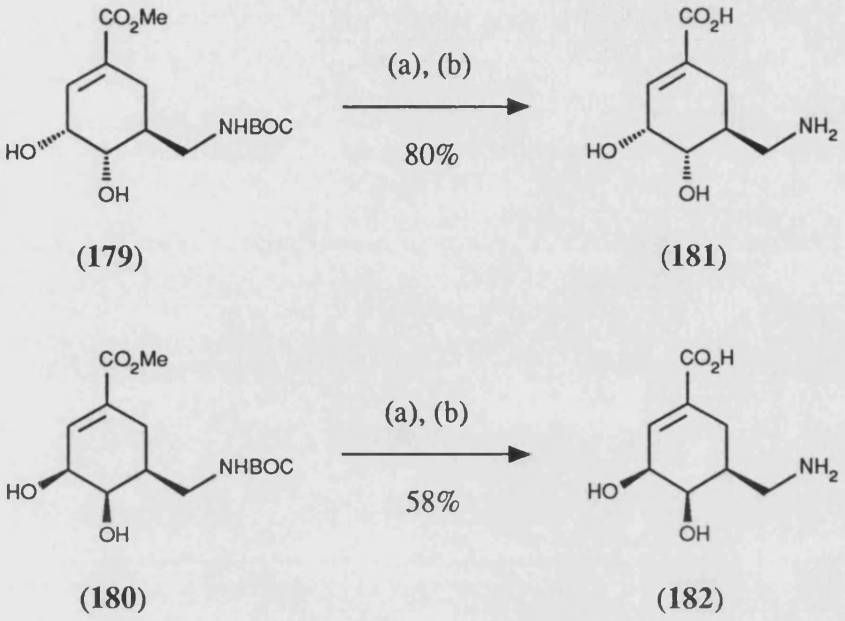
Figure 2.10 ORTEP Diagram of (**179**)

The ^{1}H NMR spectrum of the other diol, (**180**), was less informative, the resonances due to 2-H, 3-H and 4-H all appearing as broad singlets. Nevertheless, the spectrum showed great similarity to those of previous compounds with (3β, 4β, 5β) stereochemistry and the assignment is not in doubt.

2.4.2 Deprotection of the Diols (179) and (180)

The diols (**179**) and (**180**) are the protected forms of two novel amino acids, which were of interest for biological testing. Deprotection of each of these compounds was accomplished by first cleaving the *N-tert*-butoxycarbonyl group,

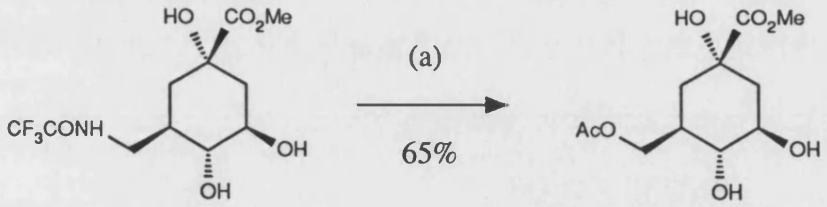
through treatment with trifluoroacetic acid (**Scheme 2.29**). The intermediate amino esters were not isolated since they were highly water soluble and thus extraction was difficult. Instead, the crude products were treated with aqueous sodium hydroxide solution to effect saponification and the amino acids (**181**) and (**182**), were subsequently purified by ion-exchange chromatography.



Scheme 2.29 Reagents: (a) TFA, 20°C, 5 min; (b) aq. NaOH, H_2O, 20°C, 20 h.

2.4.3 Deamination of the diol (179)

A different deamination procedure was utilised for the diol (**179**), instead of using a disulphonimide. Our attention was drawn to the conversion of aliphatic amines to alcohols, *via* *N*-nitrosation of the corresponding amides, by a recent publication by Ganem.[85] As part of a synthesis of a shikimate pathway enzyme inhibitor,[86] the quinate derived trifluoroacetamide (**183**) was transformed into the acetate (**184**) by treatment with sodium nitrite in acetic acid and acetic anhydride (**Scheme 2.30**). The acetate was obtained in 64% yield. Elimination, rearrangement or esterification of the free hydroxyl groups, was not reported.

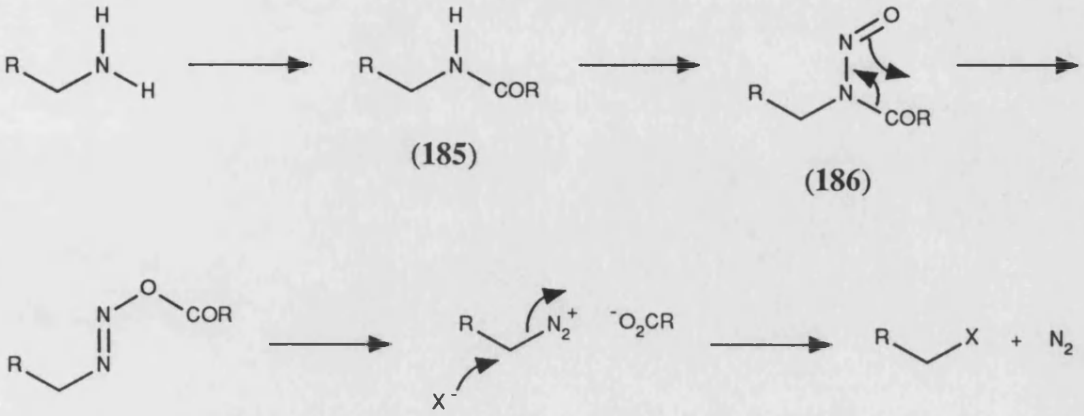


(183) (184)

Scheme 2.30 Reagents: (a) $NaNO_2$, AcOH, Ac_2O, 0°C.

The deamination of aliphatic amines *via* conversion into their *N*-nitroso-amides has been widely studied.[87] The conventional procedure involves nitrosation of an amide (**185**) to give a *N*-nitrosoamide (**186**), which on heating rearranges to a diazoester (**187**) (**Scheme 2.31**). This transient intermediate cannot be isolated, but rapidly breaks down to afford a diazonium salt (**188**), the cation of which can either react with the carboxylate counter anion to form an ester, or react with solvent to give a solvolysis product or undergo an elimination reaction to yield an olefin.

Rearrangement of *N*-nitrosotrihaloacetamides is more rapid than that of simple *N*-nitrosoacetamides and in Ganem's procedure[85] the reaction is complete at 0°C; no intermediates are observed and if acetic acid is present in the solvent mixture, the diazonium species (**188**) affords the corresponding acetate.

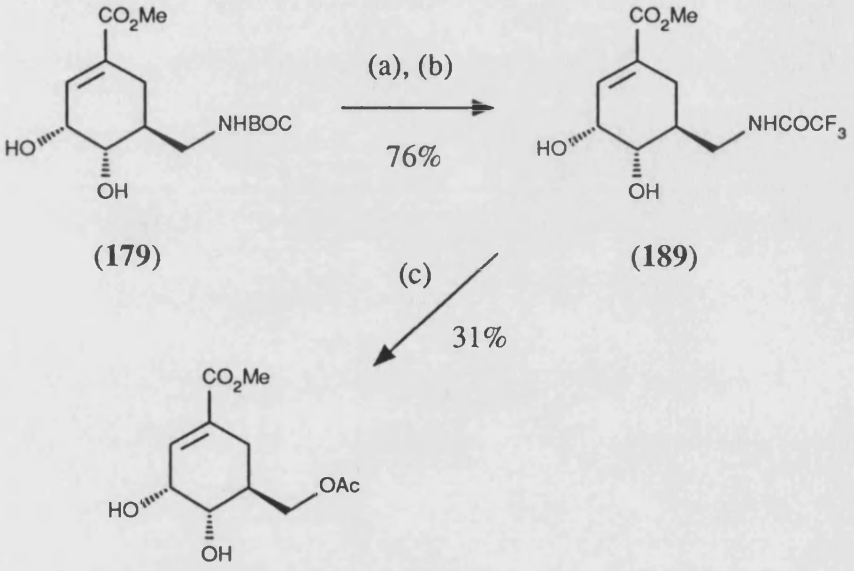


(185) (186) (187) (188)

Scheme 2.31

In order to apply this procedure to our compound, the diol carbamate (**179**) was converted into the trifluoroacetamide (**189**) (**Scheme 2.32**). A 'one-pot' procedure was used, treatment with trifluoroacetic acid cleaved the *tert*-butoxycarbonyl group and the crude product was then acylated using trifluoroacetic anhydride in pyridine, to afford the trifluoroacetamide (**189**) in 76% yield.

Nitrosation of this compound was performed according to Ganem's conditions, starting at -4°C initially and allowing the reaction mixture to warm to 10°C overnight. The acetate (**190**) was isolated in only 31% yield, although this was the major product. From TLC analysis, traces of other products were evident which appeared less polar than the main product, unfortunately these minor compounds could not be obtained pure.



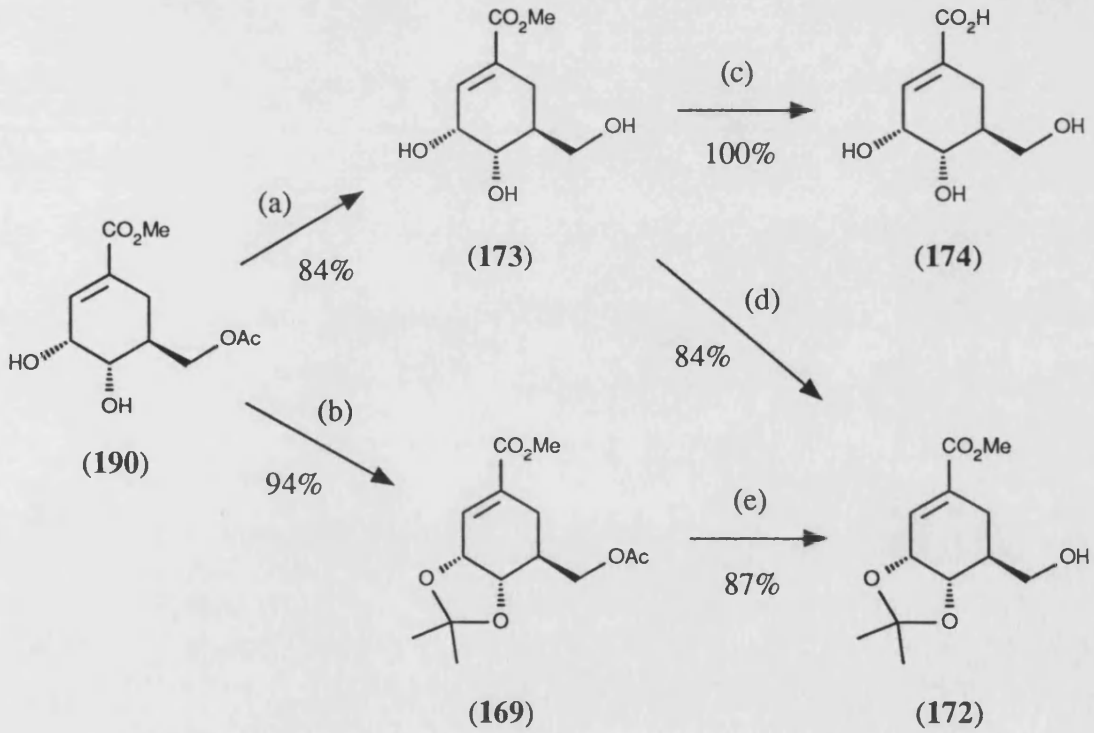
Scheme 2.32 Reagents: (a) TFA, 20°C, 30 min; (b) TFAA, Py, DMAP, 20°C, 3 days; (c) $NaNO_2$, AcOH, Ac_2O, -4 to 10°C, 17 h.

The reaction was repeated several times and since no unreacted starting material was observed, the low yield is most likely due to hydrolysis of the methyl ester. Although this yield was disappointing, especially in comparison to the yields

reported by Ganem for other compounds, it was felt that this was acceptable since fewer steps were involved than the previous deamination method.

2.4.4 Deprotection and Protection of the Diol Acetate (190)

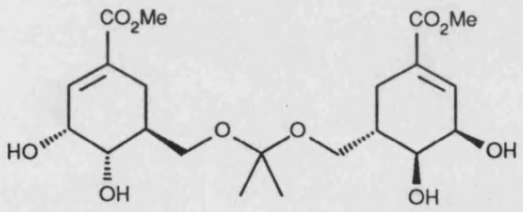
To complete the synthesis of 5-homoshikimic acid by this route, a sequence of straightforward deprotection steps were necessary (**Scheme 2.33**). Methanolysis of the acetate (**190**) yielded methyl 5-homoshikimate (**173**), identical in all respects with the sample prepared by previous routes and thus providing further confirmation of our earlier stereochemical assignments. This compound was saponified, as before, to afford 5-homoshikimic acid (**174**). The complete route to this compound involved a total of nine steps, from pyridine, representing a significant improvement over our early work.



Scheme 2.33 Reagents: (a) aq. NH_3, MeOH, 20°C, 21 h; (b) $Me_2C(OMe)_2$, Me_2CO, *p*-TSA, 20°C, 18 h; (c) NaOH, H_2O, 20°C, 5.5 h; (d) $Me_2C(OMe)_2$, Me_2CO, *p*-TSA, 20°C, 22 h; (e) aq. NH_3, MeOH, 20°C, 48 h;

The acetate (**190**) was also subjected to further protection/deprotection reactions, in order to access the acetonide (**172**) which was required for some further studies (**Section 2.5**). Protection of the two hydroxyl groups yielded the previously prepared acetonide acetate (**169**), which upon methanolysis afforded the acetonide (**172**), as before.

Preparation of this acetonide (**172**) was also investigated *via* the triol (**173**), however, the overall yield was lower. This was due to the formation of the ketal (**191**) as a by-product, in the conversion of the triol (**173**) to the acetonide (**172**). The structure of the ketal (**191**) was deduced from examination of the ^{1}H and ^{13}C NMR spectra and supported by mass spectrometric data. Interestingly, the compound is symmetric and the ^{1}H NMR spectrum for each half should be identical. However, since the compound was formed from a racemic triol (**173**), two diastereomeric (and racemic) compounds are possible. This was evident from the complex nature of ^{1}H NMR spectrum and the duplication of signals in the ^{13}C NMR spectrum.

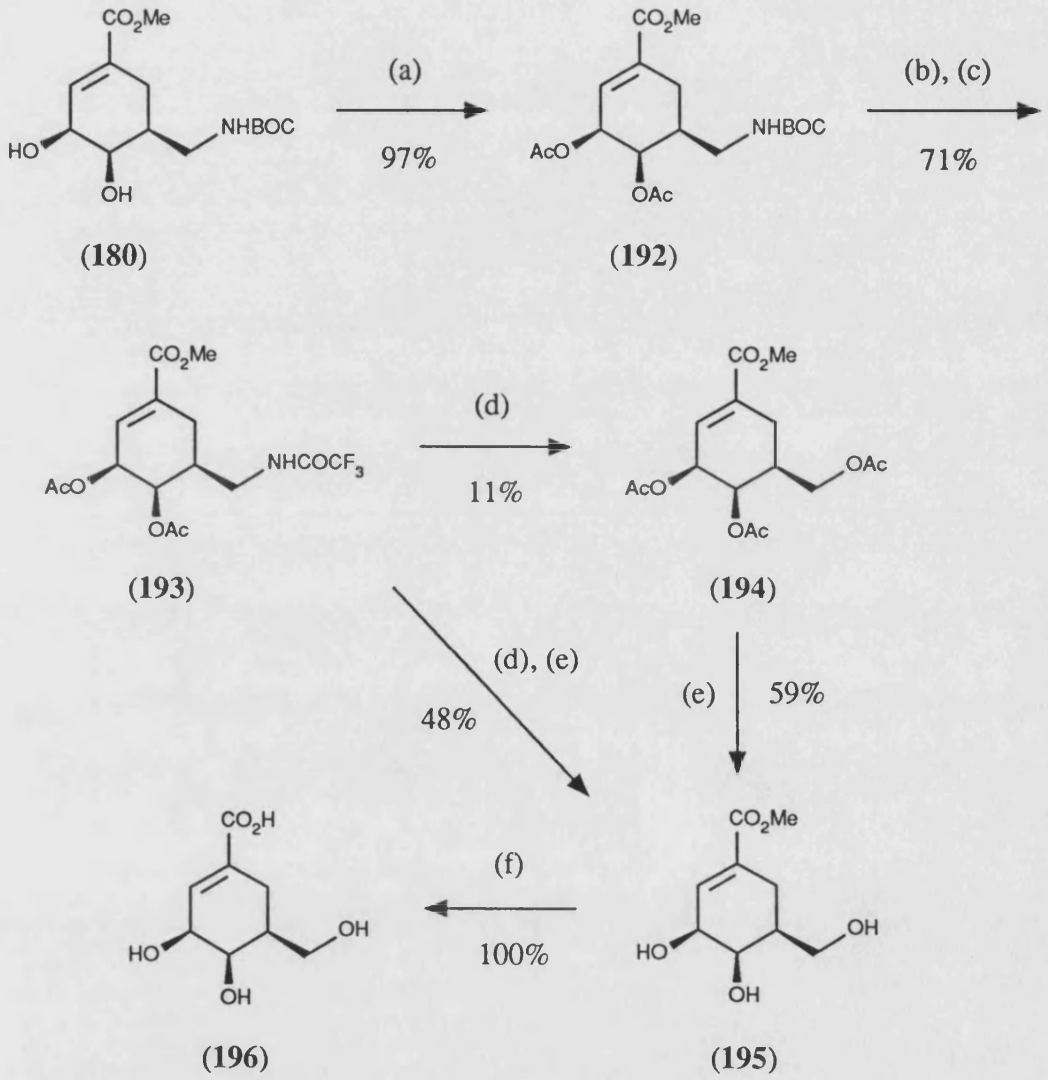


(191)

2.4.5 Synthesis of 5-*epi*-Homoshikimic acid

Since the osmium tetroxide catalysed *cis*-hydroxylation of the diene (**135**) produced both the (3β, 4β, 5β) diol (**180**) as well as the (3α, 4α, 5β) diol (**179**), the triol with (3β, 4β, 5β) stereochemistry was potentially just as accessible as 5-homoshikimic acid. The deamination - deprotection sequence was therefore applied to compound (**180**), in order to prepare a further derivative for biological assessment.

The diol (**180**) was first converted to the diacetate (**192**), by standard methods, in order to investigate whether this would lead to an improved yield for the deamination step (**Scheme 2.34**). This also served to purify the starting material, the diol (**180**) being non-crystalline and slightly difficult to obtain pure. The diacetate (**192**) was then converted to the trifluoroacetamide (**193**), using the same procedure as before.



Scheme 2.34 Reagents: (a) Ac_2O, Et_3N, DMAP, 20°C, 21 h; (b) TFA, 20°C, 10 min; (c) TFAA, Py, DMAP, 20°C, 1.5 h; (d) $NaNO_2$, AcOH, Ac_2O, -4°C to 0°C, 18 h; (e) aq. NH_3, MeOH, 20°C, 2 days; (f) NaOH, H_2O, 20°C, 18 h.

Nitrosation of this trifluoroacetamide gave a complex mixture, from which the desired triacetate (**194**) could be separated by chromatography. The yield of this product was very poor and TLC analysis indicated several minor products at higher R_F than the triacetate, as well as some more intense spots at lower R_F. Methanolysis of the triacetate (**194**) afforded the triol (**195**).

It was anticipated that the lower R_F products observed in the nitrosation reaction were various hydroxy acetates, produced by hydrolysis of one or more of the acetate groups during the reaction. This was proved by performing the nitrosation reaction and then carrying out the methanolysis on the crude product, before any chromatographic purification. The major product produced from this sequence of reactions was indeed the triol (**195**), obtained in 48% yield. As before, a few minor products were evident by TLC, these were not characterised. This two step procedure proved far more convenient than separation of the triacetate.

Saponification of the triol (**195**) yielded the acid (**196**), (±)-5-*epi*-homo-shikimic acid. The ^1H NMR spectrum of this compound was not particularly informative, in contrast to that of 5-homoshikimic acid. The resonances due to 2-H, 3-H and 4-H appeared as unresolved multiplets or broadened singlets. However, the similarity of the spectrum to those of the previously prepared compounds with (3β, 4β, 5β) stereochemistry was clear and it is likely that 5-*epi*-homoshikimic acid adopts the same solution conformation (see page 20).

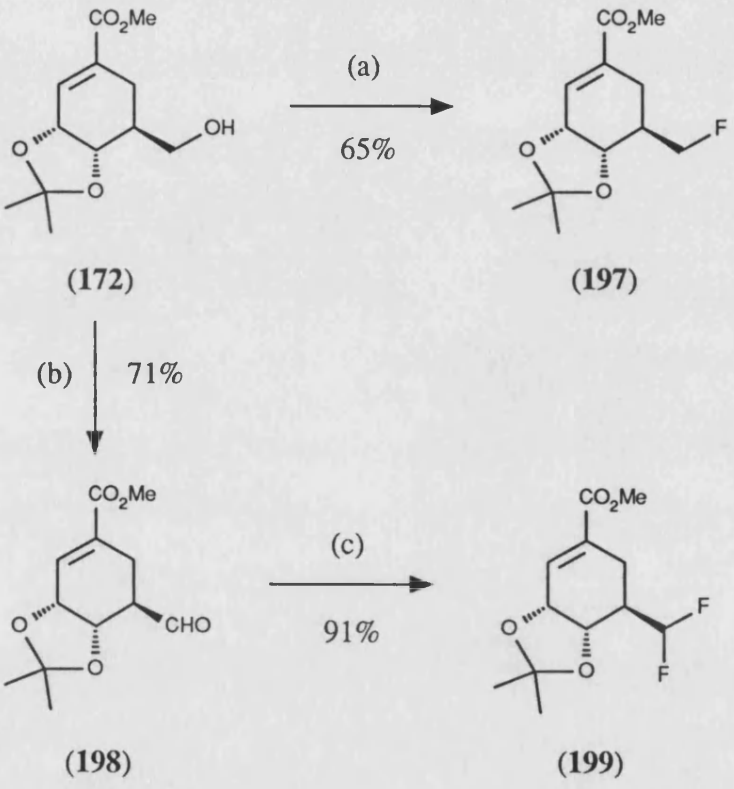
2.5 Synthesis of 5-Fluoromethyl Shikimate Analogues

Replacement of a hydroxyl group by a fluorine atom is a frequent ploy used by medicinal chemists seeking to modify biological processes. Since some of the homoshikimates show interesting bioactivities, we considered it important to prepare the 5-fluoromethyl analogues of shikimic acid.

2.5.1 Fluorination

The conversion of oxygen-containing functional groups to fluorinated substituents is commonly achieved using analogues of sulphur tetrafluoride. Diethylaminosulphur trifluoride (DAST)[88] has been widely used, however, recently morpholinosulphur trifluoride (morph-DAST)[89] has been reported as an alternative. The latter reagent is safer to use than DAST and reportedly gives higher yields of fluorinated products.

Treatment of the previously described alcohol (**172**) with morph-DAST in dichloromethane afforded the fluoride (**197**) in 65% yield, with 23% recovered starting material (corrected yield 85%) (**Scheme 2.35**).



Scheme 2.35 Reagents: (a) morph-DAST, CH_2Cl_2, -78°C to 20°C, 2 days; (b) PCC, CH_2Cl_2, 20°C, 4 h; (c) morph-DAST, CH_2Cl_2, 20°C, 2 days.

In order to obtain the difluoro analogue, it was first necessary to oxidise the

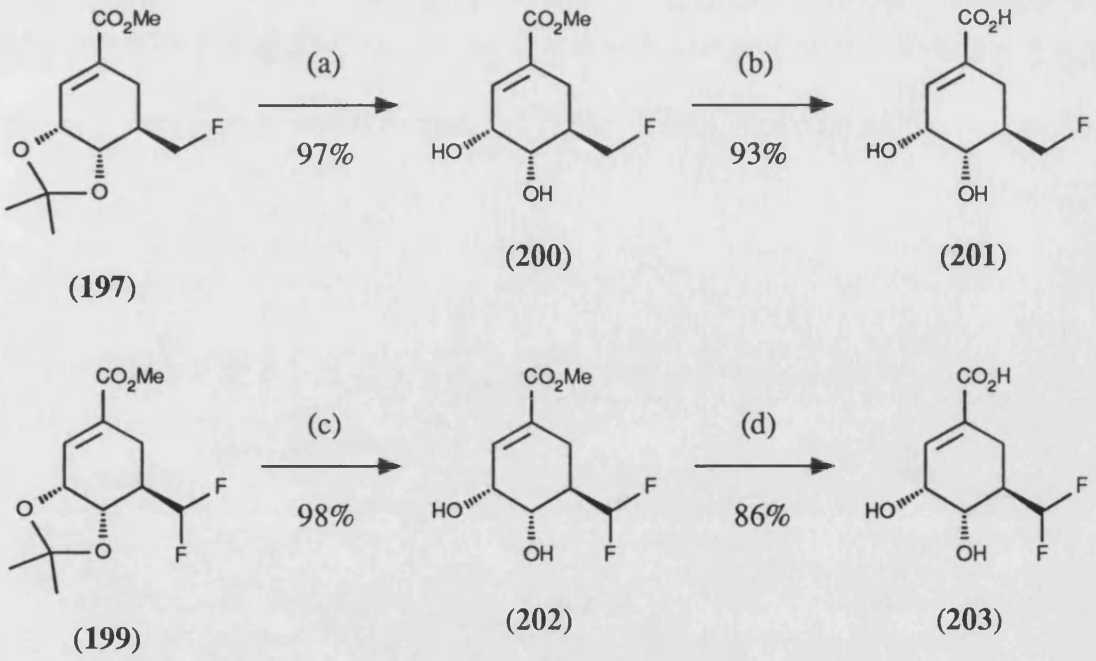
alcohol to the corresponding aldehyde (**198**). Pyridinium chlorochromate[90] was the reagent of choice and this effected clean oxidation to afford the aldehyde (**198**) in 71% yield. A more modern reagent, tetra *n*-propylammonium perruthenate (TPAP)[91] was also tried, but found to be less convenient and lower yielding, at least on a small scale.

The structure of the aldehyde was obvious from examination of the ^{1}H NMR spectrum, with the aldehyde proton resonating as a fine doublet at δ 9.79 (J 0.5 Hz). The appearance of the resonance due to 5-H was initially puzzling: it is a broadened four line signal. A comprehensive series of homonuclear decoupling experiments, (tabulated in **Section 3.2**), established this resonance to be an overlapping doublet of doublet of doublets ($J_{5,6\beta}$ 7.0, $J_{5,4}$ 6.0, $J_{5,6\alpha}$ 5.7 Hz), with the fine coupling to the aldehyde proton unresolved. These NMR experiments also confirmed the fine allylic ($J_{6\alpha,2}$ 1.5, $J_{6\beta,2}$ 1.6 Hz) and homoallylic ($J_{6\alpha,3}$ 1.5, $J_{6\beta,3}$ 1.6 Hz) coupling present.

Fluorination of the aldehyde (**198**) with morph-DAST, afforded the difluoride (**199**) in excellent yield. These fluorination reactions were found to benefit from the long reaction times (typically 2 days), premature work-up led to reduced yields. This was not immediately obvious by TLC analysis of the reaction mixture, since the disappearance of starting material was virtually instantaneous.

2.5.2 Deprotection

Deprotection of both fluorinated products was accomplished by previously established methods (**Scheme 2.36**). Cleavage of the isopropylidene group from the fluoride (**197**) using aqueous acetic acid, afforded the diol (**200**), which was subsequently saponified with aqueous sodium hydroxide, to yield the acid (**201**). In a similar way, the difluoride (**199**) was converted *via* the diol (**202**), to the acid (**203**).



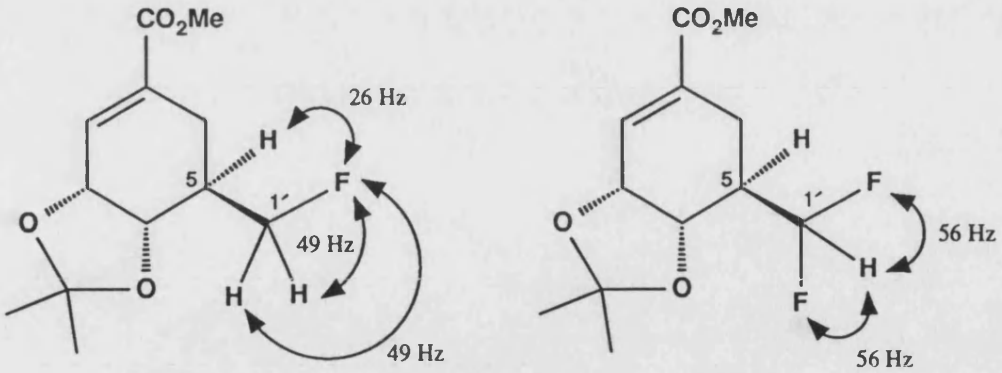
Scheme 2.36 Reagents: (a) 55% aq. AcOH, THF, 60°C, 2 days; (b) NaOH, H_2O, 20°C, 3 h; (c) 55% aq. AcOH, THF, 60°C, 3 days; (d) NaOH, H_2O, 20°C, 5 h.

2.5.3 NMR Features of the Fluorinated Products

The NMR spectra of the fluorinated products were particularly interesting, exhibiting typical H,F- and C,F-coupling. The H,F-coupling observed in the ^1H NMR spectra of the fluoride (**197**) and difluoride (**199**), is illustrated in **Figure 2.12**. The H,F-coupling for H-5 could not be determined for the difluoride (**199**), this resonance appearing as a complex multiplet.

The presence of the fluorine atom in these structures was also evident from the chemical shifts of the 1′-H resonances, these being further downfield than in earlier compounds. In the fluoride (**197**) the two 1′-H gave rise to resonances at δ 4.54 and δ 4.59, whereas the 1′-H of the difluoride (**199**) produced a large triplet of doublets at δ 6.02.

Coupling to fluorine was also observed in the ^{13}C NMR spectra, with several resonances appearing as either doublets or triplets, as appropriate (**Table 2.2**). The deprotected compounds (**200**) to (**203**) also showed similar NMR features.



(197) (199)

Figure 2.12. H,F-Coupling Constants for Compounds (**197**) and (**199**)

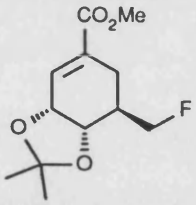
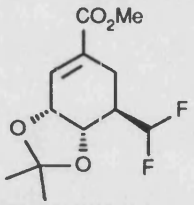
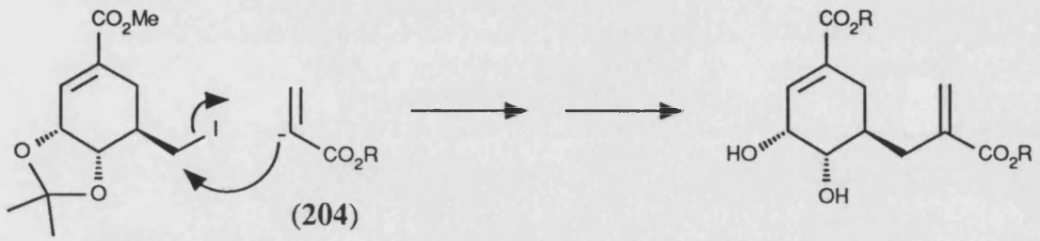
Carbon Resonance	Chemical Shift (δ)	
	 (**197**)	 (**199**)
C-1	133.1	132.4
C-2	133.7	133.3
C-3	72.9 (d, $J_{C,F}$ 4 Hz)	72.0 (d, $J_{C,F}$ 7 Hz)
C-4	71.1	70.9
C-5	38.3 (d, $J_{C,F}$ 18 Hz)	41.4 (t, $J_{C,F}$ 20 Hz)
C-6	24.4 (d, $J_{C,F}$ 4 Hz)	19.7 (t, $J_{C,F}$ 5 Hz)
C-1′	83.7 (d, $J_{C,F}$ 170 Hz)	116.0 (t, $J_{C,F}$ 241 Hz)
Other	25.7 (Me), 28.1 (Me) 52.1 (OMe), 109.2 (CMe_2) 166.6 (C=O)	25.5 (Me), 27.9 (Me) 52.1 (OMe), 109.5 (CMe_2) 166.2 (C=O)

Table 2.2 ^{13}C NMR Data for Compounds (**197**) and (**199**)

2.6 Synthesis of an 5-Enolpyruvylshikimic Acid Analogue

In order to construct analogues of later intermediates in the shikimate pathway, it was necessary to elaborate the 5-substituent of our 5-homoshikimates. It was envisaged that this could be accomplished through the reaction of the previously prepared iodide (**165**), with, formally speaking, the α-anion of a suitable acrylate (**204**) (**Scheme 2.37**).

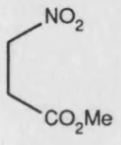


(**165**) (**204**) (**124**)

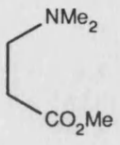
Scheme 2.37

2.6.1 α-Acrylate Anion Equivalent Reactions

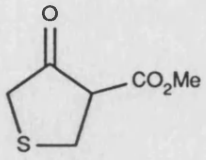
Since it has not been possible to generate an α-acrylate anion (**204**) directly, a number of synthetic equivalents have been developed.[92] The more recent α-acrylate synthons reported include: the dianion of methyl 3-nitropropanoate (**205**)[93]; the anion of methyl 3-(dimethylamino)propanoate (**206**)[94] and the anion of 4-methoxycarbonylthiolan-3-one (**207**).[95] In each case α-alkylation, followed by β-elimination, yields the required α-alkylated acrylate.



(**205**)



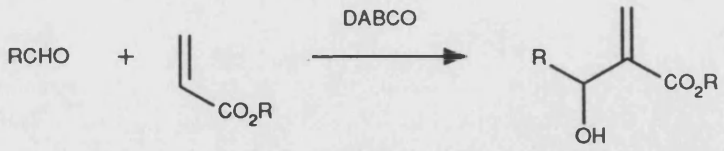
(**206**)



(**207**)

A related reaction uses the DABCO-catalysed coupling of an aldehyde with an acrylic ester, to form 2-(hydroxyalkyl)acrylic esters (**Scheme 2.38**).[96] This

approach is not suitable for alkyl halides however, and addition transformations are required to form the de-hydroxylated product.[97]

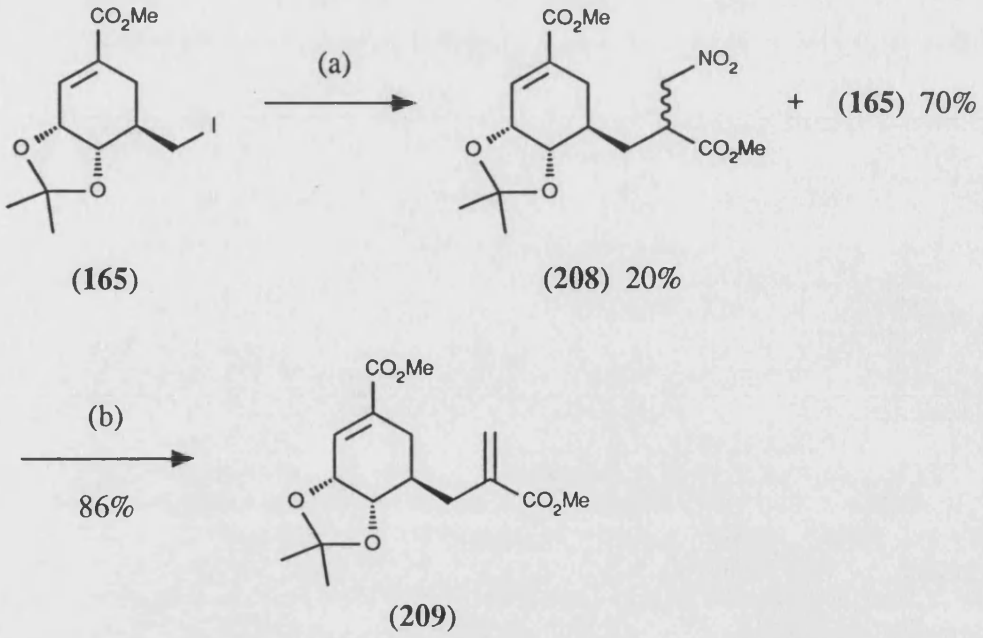


Scheme 2.38

For our attempted synthesis, Seebach's approach,[93] using the dianion of methyl 3-nitropropanoate (**205**), was chosen. This reagent seemed the most convenient, the starting material being commercially available and a single elimination step being involved. Treatment of (**205**) with two equivalents of LDA, was carried out in a 2:1 mixture of THF - DMPU, according to Seebach's procedure. The co-solvent DMPU is necessary in order to solubilise the dianion and was preferred to the alternative, but carcinogenic, solvent HMPA. Reaction of this dianion with the iodide (**165**) afforded the nitro compound (**208**), as a mixture of diastereomers, in only 20% yield (**Scheme 2.39**). The bulk of the material recovered was unreacted starting material (**165**).

Further attempts to optimise this reaction were unsuccessful. A slight turbidity of the dianion solutions was observed, however, sonication of the reaction mixture in an ultrasound bath, did not improve the product yield. The poor result was attributed to unreactivity of the iodide (**165**), since, although primary, this iodide is almost as sterically hindered as a neopentyl iodide.

Full characterisation of the nitro compound (**208**) was not possible due to the inseparable mixture of diastereomers obtained. Nonetheless, the presence of the side chain was evident from the IR spectrum, which contained two distinct carbonyl bands at 1735 (C=O, saturated ester) and 1715 (C=O, unsaturated ester) and from the mass spectrum which showed a molecular ion at m/z 258 (MH^+, 31%).

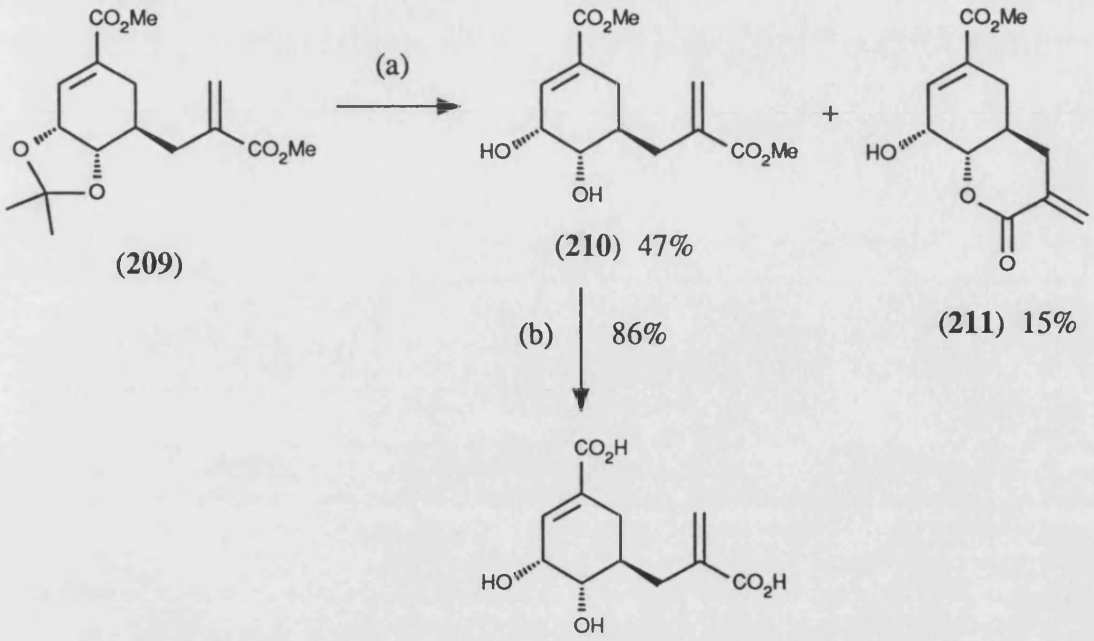


Scheme 2.39 Reagents: (a) $NO_2(CH_2)_2CO_2Me$, 2 equiv. LDA, THF, DMPU, -78 to 0°C, 13 h; (b) DBU, THF, 20°C, 4 h.

Treatment of (**208**) with the base 1,8-diazabicyclo[5.4.0]undec-7-ene, effected a clean elimination of nitrous acid, to give the dialkene (**209**) in 86% yield (**Scheme 2.39**). The presence of the exocyclic double bond was apparent from the ^1H NMR spectrum, which showed two broad singlets, exhibiting fine allylic coupling, at δ 5.56 and 6.24. Full assignment was facilitated by homonuclear decoupling experiments (tabulated in **Section 3.2**).

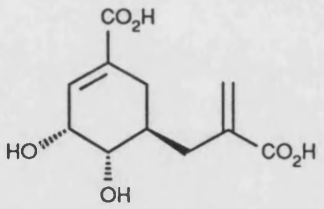
2.6.2 Deprotection

Deprotection of the dialkene (**209**) followed the same procedure as for previous compounds (**Scheme 2.40**). Removal of the isopropylidene group, using aqueous acetic acid, yielded the diol (**210**) and also effected cyclisation, to give a small amount of the bicyclic lactone (**211**). The two compounds were separated by chromatography and the diol (**210**) saponified to afford the diacid (**212**), which is the carba analogue of 5-enolpyruvylshikimic acid (**70**).

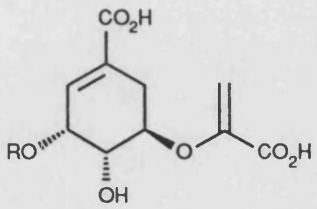


Scheme 2.40 Reagents: (a) 50% aq. AcOH, THF, 60°C, 36 h; (b) NaOH, H_2O, 20°C, 4 h.

The structure of the lactone (**211**) is particularly interesting since it provides a means of differentiation between the C-3 and C-4 hydroxyl groups. Phosphorylation of the 3-hydroxyl group would provide access to the carba analogue of 5-enolpyruvylshikimate-3-phosphate (**8**). Very similar bicyclic lactones were used in the syntheses of 5-EPS-3P by both Ganem and Bartlett (see **Section 1.3.2**).



(**211**)



(**8**) R = H
(**70**) R = PO_3H_2

Further optimisation of this route and investigation of the chemistry of the lactone (**210**), was precluded by the development of an alternative route, from (-)-shikimic acid, within the Bath group.[98]

2.7 A Chiral Approach - Asymmetric Diels-Alder Reaction

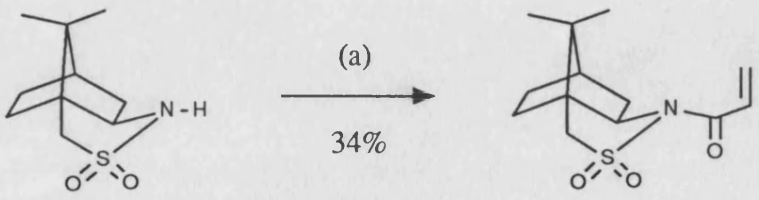
All of the compounds prepared so far, have been obtained in racemic form. The preparation of potential pharmaceutical products in homochiral form, is of the utmost importance and hence an asymmetric synthesis was sought.

The introduction of chirality through an asymmetric Diels-Alder reaction was the obvious choice, this being possible at the outset of the synthesis. High levels of asymmetric induction have been achieved in the Diels-Alder reaction, through the use of various chiral auxiliaries[99] attached to the diene or dienophile, or the employment of chiral Lewis acid catalysts.[100]

In this work, the use of an acrylate bearing Oppolzer's bornane 10,2-sultam chiral auxiliary,[101] was chosen. Previous studies[102] in this laboratory, had concentrated on chiral auxiliaries developed by Evans[103] and Helmchen,[104] with only moderate success.

2.7.1 Preparation of the Chiral Acrylate

The chiral acrylate (**214**) was prepared from the commercially available sultam (**213**), according to Oppolzer's procedure[105] (**Scheme 2.41**). The low yield of the *N*-acryloyl derivative (**214**), was similar to that recently reported for this reaction, by Kim and Lee.[106] These workers noted that reverse addition of the anion of (**213**) to acryloyl chloride, reduced polymerisation, and improved the yield, although this was still only 53%.



(**213**) (**214**)

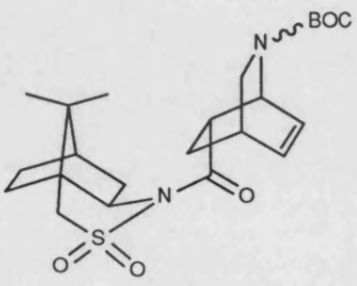
Scheme 2.41 Reagents: (a) NaH, CH_2=$CHCO_2Me$, PhMe, 20°C, 4 h.

2.7.2 Lewis-Acid Catalysed Diels-Alder Reaction

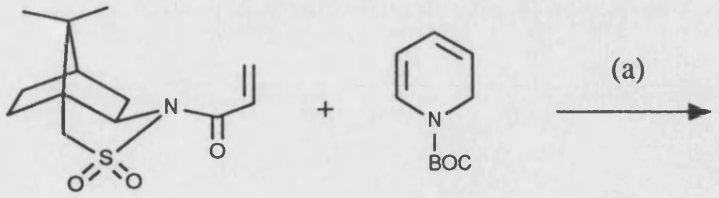
High levels of asymmetric induction in Diels-Alder reactions using chiral auxiliaries, usually require the use of a Lewis acid catalyst. The role of the catalyst is two-fold: firstly, co-ordination to the dienophile lowers the energy of the dienophile LUMO, hence decreasing the energy separation between the diene HOMO and thereby facilitating the reaction. Secondly, this co-ordination to the dienophile restricts rotation such that, ideally, approach of the diene from one face of the double bond is favoured.

A number of different Lewis acids were tried, in the reaction between the chiral acrylate (**214**) and 1-*tert*-butoxycarbonyl-1,2-dihydropyridine (**132**) (**Scheme 2.42, Table 2.3**). These reactions were conveniently monitored by TLC analysis, both starting materials being well separated from each other and also the expected Diels-Alder adducts. The position of the latter on the TLC plate was known by comparison with an authentic sample, obtained from the thermal cycloaddition reaction (discussed in the following section).

Zinc iodide was found to be effective in catalysing the reaction, (entries 2 and 3), although only a small amount of the Diels-Alder adduct was formed. Decomposition of the 1,2-dihydropyridine (DHP) was observed and thus, even though this reagent was used in a two-fold excess, it is exhausted before all of the chiral acrylate has reacted. The isolated yield of the adduct obtained in this way was only 11%, although examination of the ^{1}H NMR spectrum revealed only one diastereomer, the *endo* adduct (**216**).



(216)



(214) **(132)**

Scheme 2.42 Reagents: (a) Lewis acid and solvent as detailed in **Table 2.3**, all reactions used 0.15 mmol of (**214**), 0.30 mmol (**132**) in 3 cm^3 solvent at 20°C.

Entry	Lewis Acid	Solvent	TLC Observation
1	ZnI_2 (1.0 equiv.)	PhMe	Some adduct formation, decomposition of DHP
2	ZnI_2 (1.0 equiv.)	CH_2Cl_2	Some adduct formation, decomposition of DHP
3	ZnI_2 (3.0 equiv.)	CH_2Cl_2	Some adduct formation, decomposition of DHP
4	$ZnBr_2$ (1.0 equiv.)	CH_2Cl_2	Trace of adduct formation, decomposition of DHP
5	$MgBr_2$ (1.0 equiv.)	CH_2Cl_2	Trace of adduct formation, rapid decomposition of DHP
6	$Ti(O^iPr)_4$ (1.0 equiv.)	CH_2Cl_2	No reaction, DHP remains
7	$TiCl(O^iPr)_3$ (1.0 equiv.)	CH_2Cl_2	No reaction, DHP remains
8	None	H_2O	No reaction, DHP remains
9	aq. $LiClO_4$ (2.5M)	H_2O	No reaction, DHP remains

Table 2.3

The data for this adduct were identical with that for a sample obtained *via* the thermal cycloaddition, the stereochemistry of which was later deduced (detailed in following section).

The zinc iodide catalysed reaction was not considered synthetically useful, in view of the poor yield and other reactions, using different Lewis acids, were similarly disappointing. Two opposing effects were noticeable - using more reactive Lewis acids led to rapid decomposition of the dihydropyridine (entries 4 and 5), whereas milder Lewis acids did not promote cycloaddition at all. The latter case was exemplified by the use of titanium isopropoxides (entries 6 and 7), these Lewis acids being milder than titanium tetrachloride which has previously been shown to effect rapid decomposition of the dihydropyridine.[102]

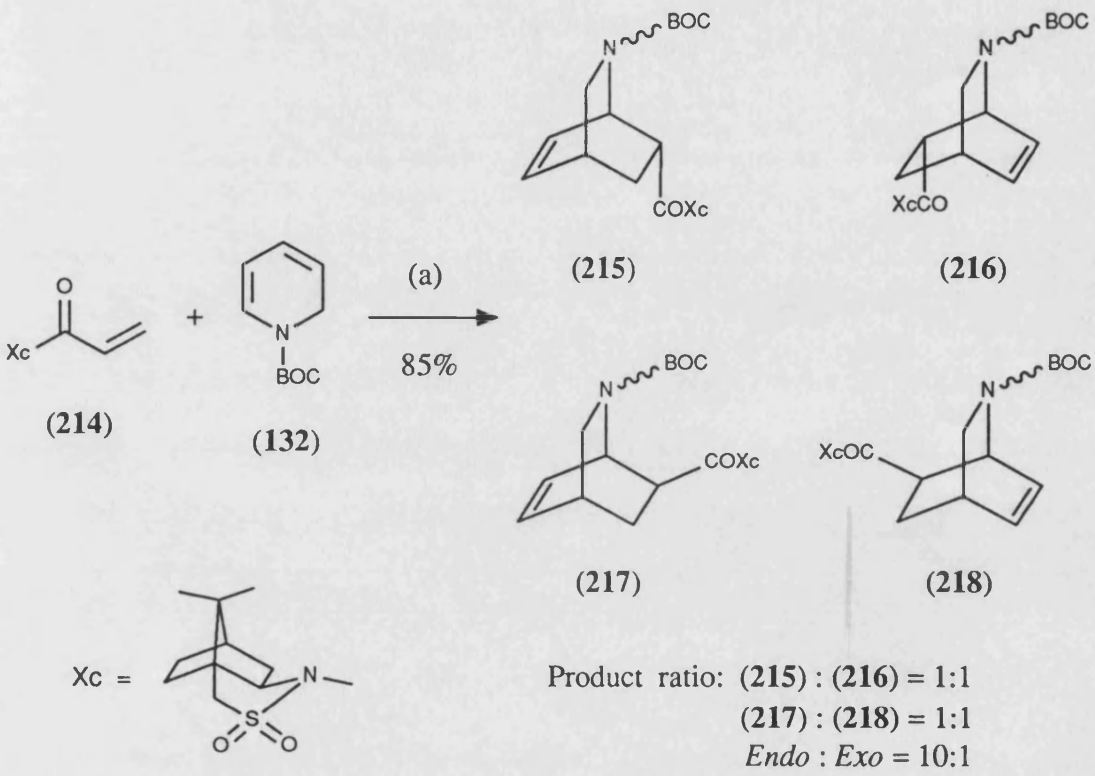
These reactions were not intended to be comprehensive and merely illustrate the problems involved in using a dihydropyridine (**132**) bearing a labile protecting group. A more suitable dihydropyridine would be 1-methoxycarbonyl-1,2-dihydropyridine (**125**), however, this would then necessitate further protection/deprotection steps later in our synthesis. Further reactions were considered outside the scope of this project and, in view of time constraints, our attention was concentrated on the thermal Diels-Alder reaction.

2.7.3 Thermal Diels-Alder Reaction

In contrast to the Lewis acid catalysed reactions, the Diels-Alder reaction of the chiral acrylate (**214**) and the dihydropyridine (**132**), proceeded cleanly to afford the mixed *exo* and *endo* adducts in 85% yield (**Scheme 2.43**). The mixed adducts were purified by column chromatography, particular care being taken to ensure that all fractions containing the products were collected, before analysis of the product ratio by ^{1}H NMR.

The δ 0.90-1.25 region of the ^{1}H NMR spectrum is of particular interest, since it contains only the resonances due to the two methyl groups of the chiral

auxiliary. In the spectrum of the mixed adducts, this region contained at least eight singlets, suggesting that all four possible diastereomers (**215**) to (**218**) were present (**Figure 2.13**). Four major peaks are observed, implying two major products, but further identification was not possible at this stage, due to the complexity of the spectrum.

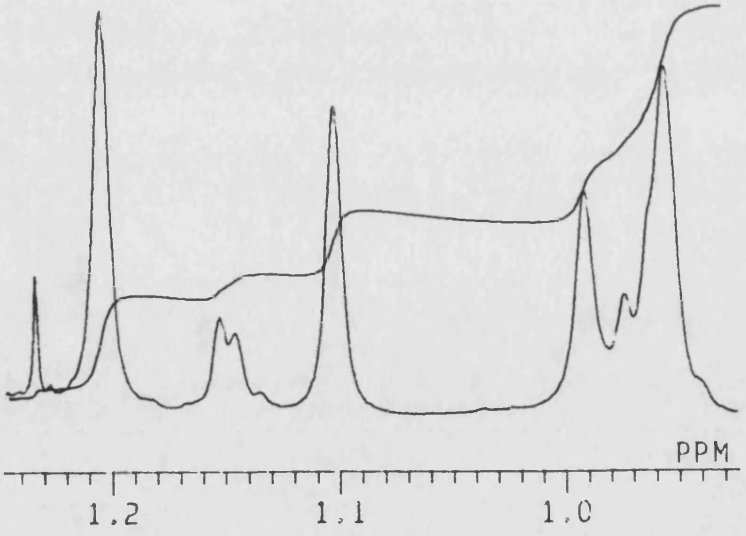


Scheme 2.43 Reagents: (a) PhMe, reflux, 2.5 days.

Recrystallisation of the mixed adducts yielded a single diastereomer, which was obviously an *endo* adduct by comparison of the ^{1}H NMR data with previous compounds. The δ 0.90-1.25 region of the spectrum for this compound, contained only two singlets, corresponding to two of the four major peaks in the spectrum of the mixed adducts (**Figure 2.13**).

Somewhat fortuitously, upon concentration and recrystallisation of the mother liquor, the other *endo* adduct was obtained. The methyl region of the ^{1}H

(a) Mixed adducts



(b) Recrystallisation
1st Crop
endo isomer (**215**)

(c) Recrystallisation
2nd Crop
endo isomer (**216**)

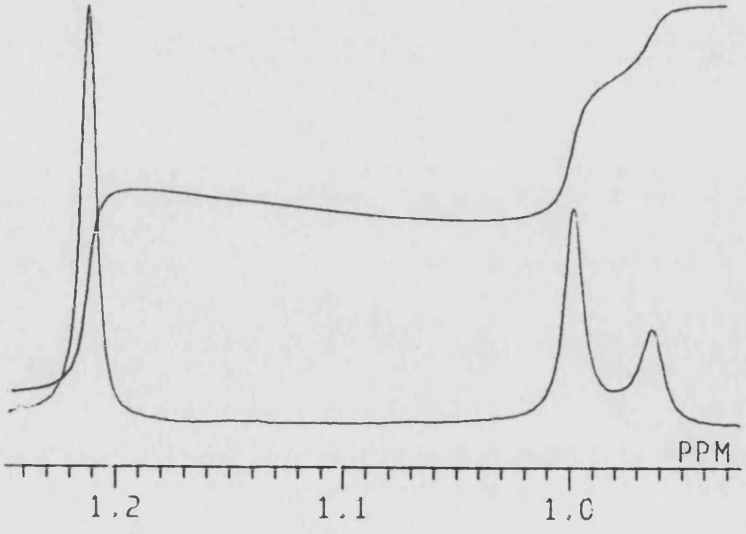


Figure 2.13

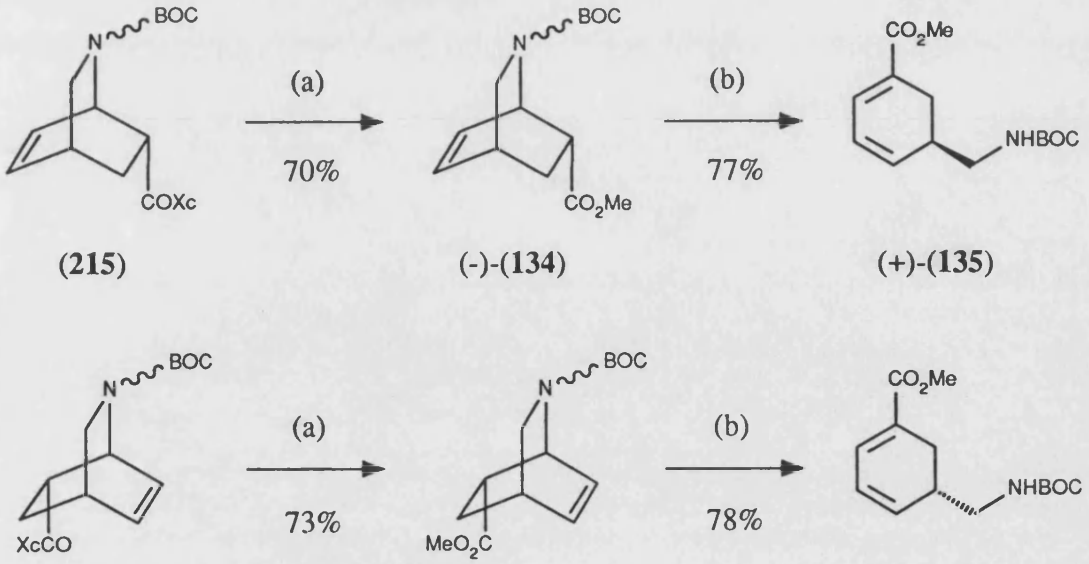
NMR spectrum for this compound contained one major singlet and a pair of smaller singlets (**Figure 2.13**). Repeated recystallisation did not alter the spectrum for this product. It was anticipated that the presence of the two smaller singlets, was due to the existence of two possible positions for the *tert*-butoxycarbonyl group attached to the nitrogen. Similar effects were noted for earlier adducts without the chiral auxiliary. In order to test this assumption the spectrum was recorded at a higher temperature (60°C). Coalescence of the two smaller singlets was observed, resulting from the increased rate of exchange at this temperature.

On the basis of these findings, it can be seen that the thermal reaction gave rise to predominantly *endo* adducts. The two possible *endo* adducts were formed in approximately 1:1 ratio and the *endo*:*exo* ratio was tentatively assigned as 10:1. The lack of asymmetric induction under these reaction conditions is not unexpected.

When the reaction was repeated on a larger scale, reasonable quantities of both *endo* adducts could be obtained. The two products were separated by repeated column chromatography and recrystallised to obtain pure materials. The diastereomeric excess (d.e.) was estimated to be greater than 98% from ^{1}H NMR measurements.

The problem of determining the absolute stereochemistry of each *endo* adduct remained and this was achieved by first removing the chiral auxiliary. Methanolysis of each adduct using potassium carbonate in methanol, afforded both enantiomers of the previously prepared compound (**134**) (**Scheme 2.44**).

The absolute configuration of each enantiomer was assigned by comparison of the specific rotations with literature values for a similar compound. Both enantiomers of the methoxycarbonyl derivative (**219**), were prepared by Marazano[107] and show very similar rotations to our compounds (**Table 2.4**). On this basis the structure of the adducts bearing the chiral auxiliary, (**215**) and (**216**), were also assigned.



Scheme 2.44 Reagents: (a) K_2CO_3, MeOH, 20°C, 2.5 h; (b) *n*-BuLi, $(TMS)_2NH$, THF, -78 to 20°C, 1 h.

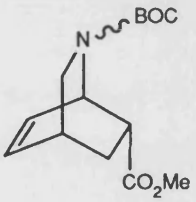
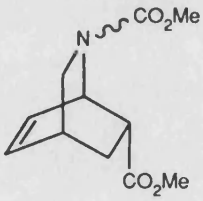
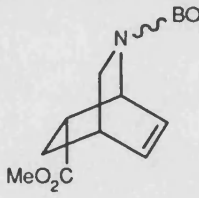
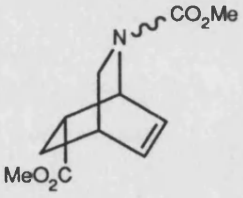
Specific Rotation $[\alpha]_D^{20}$			
			
(-)-(**134**)	(-)-(**219**)	(+)-(**134**)	(+)-(**219**)
-106° (c = 1.02, $CHCl_3$)	-110° Reference 107	+104° (c = 1.01, $CHCl_3$)	+109° Reference 107

Table 2.4

Having achieved what amounts to a resolution of the adduct (**134**), both enantiomers were ring opened in the same way as the racemic compound, to afford

the two enantiomers of the diene (**135**). The optical rotations of these compounds were also instructive and could be compared to another literature compound (**220**) prepared by Birch[28] (**Table 2.5**). Compound (**220**) was an intermediate in an asymmetric synthesis of (-)-gabaculine and both these compounds are strongly levorotatory, thus supporting our assignment of (**135**).

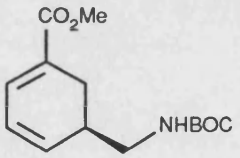
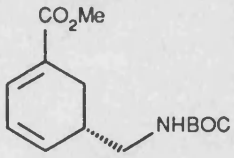
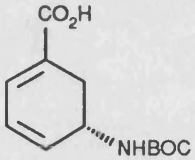
Specific Rotation $[\alpha]_D^{20}$		
 (+)-(**135**)	 (-)-(**135**)	 (-)-(**220**)
+200° ($c = 0.4$, $CHCl_3$)	-195° ($c = 0.4$, $CHCl_3$)	-249° ($c = 0.1$, $CHCl_3$)[28]

Table 2.5

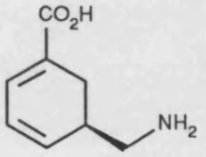
Since the racemic form of the diene (**135**), had previously been converted to 5-homoshikimic acid, preparation of both enantiomers of this compound, (and our other targets), was now possible. The amount of homochiral diene (**135**) available and lack of time, precluded completion of the sequence.

2.8 Summary

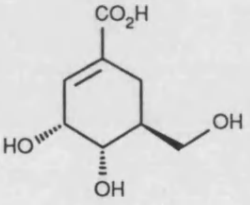
The total syntheses of (±)-5-homogabaculine (**129**) and (±)-5-homo-shikimic acid (**174**) have been described, from readily available 1,2-dihydro-pyridines. Improved syntheses of the latter have been developed, also leading to a range of analogues (**177**), (**181**), (**182**) and (**196**). In addition, the 5-fluoromethyl shikimate (**201**) and 5-difluoromethyl shikimate (**203**) have been prepared. Stereochemical and NMR features have been rationalised throughout.

The synthesis of (**212**), the carba analogue of 5-enolpyruvylshikimic acid has been detailed and a way by which the 3-hydroxyl group may be selectively phosphorylated is suggested.

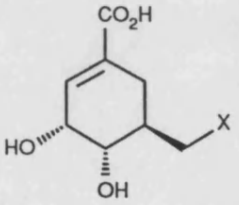
A key intermediate in the synthesis of these compounds has been prepared in homochiral form.



(129)

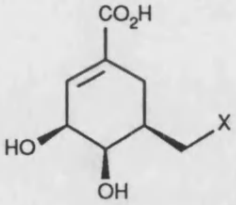


(174)



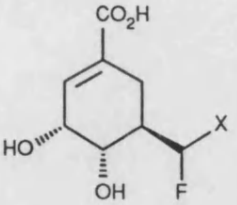
(177) X = I

(181) X = NH_2



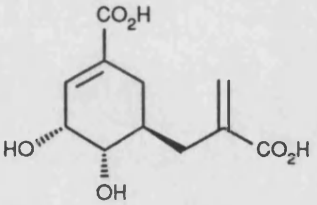
(182) X = NH_2

(196) X = OH



(201) X = H

(203) X = F



(212)

CHAPTER THREE

EXPERIMENTAL

CHAPTER THREE

EXPERIMENTAL

3.1 Instrumentation and Experimental Techniques

3.1.1 Solvents and Reagents

All solvents were dried and distilled before use. Petrol refers to petroleum ether boiling in the range 60-80°C and light petrol refers to that boiling in the range 40-60°C. Tetrahydrofuran was pre-dried over sodium wire and then refluxed over sodium benzophenone ketyl under a nitrogen atmosphere until anhydrous. This was redistilled immediately prior to use. Osmium tetroxide was used as a solution in *tert*-butanol prepared according to the procedure of Daniels and Fischer.[108] All other solvents and reagents were purified using the procedures described in *Purification of Laboratory Chemicals*.[109]

3.1.2 Chromatography

Thin layer chromatography (TLC) was used extensively as a qualitative guide during reactions and for assessing the purity of compounds. Merck DC-alufolien Kieselgel 60 F_{254} sheets containing fluorescent indicator were used for this purpose. Visualisation of compounds was acheived by illumination under short wavelength (254 nm) ultraviolet light (when possible). Plates were developed by treatment with either a 0.5% (w/v) aqueous solution of potassium permanganate (followed by washing in water) or a 7% (w/v) methanolic solution of phosphomolybdic acid, followed by warming of the TLC plate.

Medium pressure flash column chromatography was routinely employed using Amicon Matrex or Merck 9385 silica gel. Columns were packed as a slurry in the eluting solvent and the material to be chromatographed introduced directly as a

solution in the eluting solvent or preabsorbed onto the column support and then applied as a thin layer to the top of the column. A pressure gradient was developed using a small hand bellow.

3.1.3 General

Glassware used for moisture sensitive reactions was heated in an oven at 120°C overnight and then allowed to cool in a desiccator over $CaCl_2$. Flasks and stirrer bars were additionally flame dried under a stream of dry nitrogen prior to use.

Solvents were evaporated with a Büchi rotary evaporator using a water aspirator or a vacuum pump as required and a water bath temperature <40°C to avoid unnecessary heating.

All dilute aqueous solutions used were 2.0 M unless otherwise stated.

3.1.4 Analysis and Spectroscopy

Melting points (m.p.) were determined on commercially available apparatus (Electrothemal Mk III or Gallenkamp) and are uncorrected. Elemental micro-analyses were carried out using a Carlo Erba 1106 Elemental Analyser. Optical rotations were measured using a Perkin-Elmer 141 polarimeter with concentration (*c*) expressed in g/100 cm^3.

Infrared spectra were recorded in the range 4000-600 cm^{-1} using a Perkin-Elmer 1310 spectrophotometer and peaks are reported (ν_{max}) in wavenumbers (cm^{-1}). Samples were prepared as liquid films, nujol mulls or chloroform solutions, as indicated.

Proton magnetic resonance spectra were recorded on a Jeol GX FT 270 (270 MHz) spectrometer although, where indicated, Jeol GX FT 400 (400 MHz) or Varian EM-360 (60 MHz) instruments were used. Carbon-13 magnetic resonance spectra were recorded on a Joel GX FT 270 spectrometer operating at 67.8 MHz and using 90 and 135 DEPT pulse sequences to aid multiplicity determination. Chemical

shifts (δ) are expressed in parts per million downfield from internal tetramethylsilane. The multiplicities of the resonances are denoted by s (singlet), d (doublet), t (triplet), q (quartet) and m (multiplet). The abbreviation br (broadened) is used to indicate significant broadening, whether due to rapid exchange or unresolved fine coupling. Homonuclear decoupling experiments and 2D homonuclear shift correlated (COSY) spectra were used to confirm proton assignments when required.

Mass spectra were recorded using a VG Analytical 7070E instrument with a VG 2000 data system. Electron ionisation (E.I.) spectra were produced using an ionising potential of 70 eV. Chemical ionisation (C.I.) was employed using isobutane as the reagent gas, although where indicated, ammonia was also used.

3.2 Experimental Procedure

1-(Methoxycarbonyl)-1,2-dihydropyridine (125)[69]

A solution of methyl chloroformate (4.6 cm^3, 60 mmol) in diethyl ether (10 cm^3) was added slowly to a stirred suspension of sodium borohydride (2.34 g) in pyridine (4.9 cm^3, 60 mmol) and methanol (30 cm^3) at -78°C under a nitrogen atmosphere. The rate of addition was controlled to maintain the temperature below -60°C. After 2 hours the reaction mixture was poured into ice-water (*ca.* 400 cm^3), allowed to warm to 20°C and extracted with ether (4 x 80 cm^3). The combined extracts were dried ($MgSO_4$) and concentrated under reduced pressure. Chromatography on a short alumina column eluted with diethyl ether yielded the title compound as a colourless oil (5.39 g, 65%): R_F 0.78 (1:1 petrol-ethyl acetate); ν_{max}(liquid film) 1705 (C=O), 1650 (C=C), 1585, 1440, 1340, 1280, 1235, 1120, 970, 770, 700 cm^{-1}; δ_H(60 MHz, $CDCl_3$) 3.75 (3H, s, OMe), 4.30 (2H, dd, *J* 2, 3 Hz, 2 x 2-H), 4.80-6.00 (3H, m, 3-H, 4-H, 5-H), 6.70 (1H, m, 6-H).

2,7-Di-(methoxycarbonyl)-2-azabicyclo[2.2.2]oct-5-ene (126)

A solution of the dihydropyridine (125) (2.90 g, 20.9 mmol) and methyl acrylate (3.76 cm^3, 41.8 mmol) in toluene (10 cm^3) was heated to reflux under a nitrogen atmosphere. After 3 days the solvent and excess methyl acrylate was evaporated under reduced pressure. Column chromatography (petrol-ethyl acetate 4:1 to 1:1) yielded the title compound, a mixture of *exo* and *endo* adducts, as a colourless oil (3.45 g, 73%): R_F 0.58 (1:1 petrol-ethyl acetate); ν_{max}(liquid film) 1710 br (C=O), 1440, 1390, 1345, 1305, 1280, 1195, 1115 cm^{-1}; $\delta_H(CDCl_3)$ 1.57 (1H, m, 8_{exo}-H), 1.86 (2H, m, 2 x 8_{endo}-H), 2.10 (1H, d, J_{gem} 12.0 Hz, 8_{exo}-H), 2.57 (1H, m, 7_{exo}-H), 2.82 (2H, m, 4_{exo}-H, 4_{endo}-H), 2.96 (2H, m, 3_{exo}-H, 3_{endo}-H), 3.07 (1H, m, 7_{endo}-H), 3.27 (1H, d, J_{gem} 10.0 Hz, 3_{endo}-H), 3.38 (1H, dd, J_{gem} 10.0, $J_{3,4}$ 2.0 Hz, 3_{exo}-H), 3.64 (3H, s, OMe), 3.66 (3H, s, OMe), 3.69 (3H, s, OMe), 3.70 (3H, s, OMe), 4.96 (1H, m, 1_{exo}-H), 5.10 (1H, m, 1_{endo}-H), 6.34 (1H, m, 5_{endo}-H), 6.46 (3H, m, 5_{exo}-H, 6_{exo}-H, 6_{endo}-H); m/z (E.I.) 225 (M^+, 225.0994 $C_{11}H_{15}NO_4$ requires 225.1000, 2%), 194 (7), 139 (100), 124 (38).

Methyl 5-[N-(methoxycarbonyl)aminomethyl]cyclohexa-1,3-diene-1-carboxylate (127)

A solution of 1,1,1,3,3,3-hexamethyldisilazane (1.78 cm^3, 8.44 mmol) in dry THF (8 cm^3) was cooled to -78°C under a nitrogen atmosphere and a solution of *n*-butyl lithium in hexanes (1.6 M, 8.44 mmol) was added slowly. After 30 min a solution of the adducts (126) (1.81 g, 8.04 mmol) in THF (20 cm^3) was added dropwise *via* cannular. The reaction mixture was stirred for 20 min at this temperature, allowed to warm to 20°C and quenched with saturated aqueous ammonium chloride solution (20 cm^3). The mixture was extracted with dichloromethane (3 x 50 cm^3). The combined extracts were dried ($MgSO_4$) and concentrated under reduced pressure.

Column chromatography (petrol-ethyl acetate 2:1) yielded the title compound as a colourless solid (1.46 g, 81%): m.p. 51-52°C (from petrol-ethyl acetate); R_F 0.55 (1:1 petrol-ethyl acetate); (Found: C, 58.7; H, 6.8; N, 6.1. $C_{11}H_{15}NO_4$ requires C, 58.7; H, 6.7; N, 6.2%); ν_{max}(nujol) 3320 (NH), 1690 (C=O), 1530, 1430, 1265, 1250, 1225, 1180, 1085 cm^{-1}; δ_H($CDCl_3$) 2.37 (1H, ddd, J_{gem} 14.8, $J_{6\beta,5}$ 8.4, $J_{6\beta,2}$ 1.3 Hz, 6β-H), 2.59 (1H, ddd partially obscured by 5-H, J_{gem} 14.8, $J_{6\alpha,5}$ 6.4, $J_{6\alpha,2}$ 1.3 Hz, 6α-H), 2.62 (1H, m, 5-H), 3.12 (1H, m, 1′-H), 3.28 (1H, m, 1′-H), 3.67 (3H, s, OMe), 3.76 (3H, s, OMe), 4.97 (1H, br s, NH), 6.03 (1H, dd, $J_{4,3}$ 9.5, $J_{4,5}$ 3.0 Hz, 4-H), 6.13 (1H, ddd, $J_{3,4}$ 9.5, $J_{3,2}$ 5.0, $J_{3,5}$ 1.5 Hz, 3-H), 6.99 (1H, dt, $J_{2,3}$ 5.0, $J_{2,6\alpha}$ 1.3, $J_{2,6\beta}$ 1.3 Hz, 2-H); δ_C($CDCl_3$) 24.7 (C-6), 34.2 (C-5), 43.7 (C-1′), 51.6 (OMe), 52.1 (OMe), 124.8 (C-3), 126.5 (C-1), 132.6 (C-4), 134.6 (C-2), 157.1 (C=O), 167.6 (C=O); m/z (C.I.) 226 (MH^+, 33%), 194 (100), 166 (19), 137 (26).

5-[N-(methoxycarbonyl)aminomethyl]cyclohexa-1,3-diene-1-carboxylic acid **(128)**

A solution of the ester carbamate (**127**) (250 mg, 1.11 mmol) in THF (50 cm^3) was treated with aqueous sodium hydroxide solution (1.0 M, 3.33 cm^3). Water (11 cm^3) was added to give a homogeneous solution which was stirred at 20°C for 16 h. The solution was acidified with 1.0 M hydrochloric acid and extracted with ethyl acetate (3 x 50 cm^3). The combined extracts were dried ($MgSO_4$) and concentrated under reduced pressure to yield the title compound as a colourless powder (153 mg, 65%): m.p. 139-140°C (from chloroform); (Found: C, 57,2; H, 6.2; N, 6.6. $C_{10}H_{13}NO_4$ requires C, 56.9; H, 6.2; N, 6.6%); ν_{max}(nujol) 3320 (NH), 1680 br (C=O), 1620 (C=C), 1530, 1440, 1370, 1280, 1230 cm^{-1}; δ_H(D_6-DMSO) 2.12 (1H, m, 6β-H), 2.46 (2H, m, 5-H, 6α-H), 3.01 (2H, m, 2 x 1′-H), 3.53 (3H, s, OMe), 6.03 (1H, dd, $J_{4,3}$ 9.3, $J_{4,5}$ 2.4 Hz, 4-H), 6.12 (1H, dd, $J_{3,4}$ 9.3, $J_{3,2}$ 4.8 Hz, 3-H), 6.88 (1H, d, $J_{2,3}$ 4.8 Hz, 2-H), 7.30 (1H, br t, J 5.7 Hz, NH), 12.28 (1H, br s, CO_2H); δ_C(D_6-DMSO) 24.7 (C-6), 34.2 (C-5), 43.3 (C-1′), 51.4 (OMe), 124.4 (C-3), 127.4 (C-1), 131.9 (C-4),

135.3 (C-2), 157.0 (C=O), 168.1 (C=O); m/z (C.I.) 212 (MH^+, 6%), 194 (100), 180 (19), 166 (49), 162 (61), 137 (22), 123 (58).

5-(Aminomethyl)cyclohexa-1,3-diene-1-carboxylic acid (**129**)

A solution of the ester carbamate (**127**) (400 mg, 1.78 mmol) in THF (75 cm^3) was treated with aqueous sodium hydroxide solution (1.0 M, 3.55 cm^3) and water (17 cm^3). The reaction mixture was heated to reflux under a nitrogen atmosphere for 3 days, acidified with dilute aqueous hydrochloric acid and extracted with ethyl acetate (3 x 50 cm^3). The combined organic extracts were dried ($MgSO_4$) and concentrated under reduced pressure to yield the previously prepared acid (**128**) (210 mg, 56%). The aqueous phase was lyophilised to leave a white solid which was added to the top of a column of ion-exchange resin (Dowex 50/8-100, previously regenerated with dilute aqueous hydrochloric acid and packed as an aqueous slurry). The column was eluted with water to remove impurities and the product removed from the column by elution with dilute aqueous ammonia solution. Evaporation under reduced pressure and lyophilisation yielded the title compound as a cream-coloured solid (101 mg, 37%): R_F 0.70 (reverse phase silica, water); $\delta_H(D_2O)$ 2.34 (1H, ddd, J_{gem} 17.5, $J_{6\beta,5}$ 8.5, $J_{6\beta,2}$ 1.0 Hz, 6β-H), 2.56 (1H, ddd, J_{gem} 17.5, $J_{6\alpha,5}$ 8.5, $J_{6\alpha,2}$ 2.0 Hz, 6α-H), 2.71 (1H, m, 5-H), 2.93 (1H, dd, J_{gem} 12.8, $J_{1',5}$ 7.0 Hz, 1′-H), 3.01 (1H, dd, J_{gem} 12.8, $J_{1',5}$ 6.6 Hz, 1′-H), 5.95 (1H, dd, $J_{4,3}$ 9.5, $J_{4,5}$ 4.4 Hz, 4-H), 6.23 (1H, ddd, $J_{3,4}$ 9.5, $J_{3,2}$ 5.5, $J_{3,5}$ 1.5 Hz, 3-H), 6.69 (1H, br d, $J_{2,3}$ 5.5 Hz, 2-H); $\delta_C(D_2O)$ 25.3 (C-6), 31.2 (C-5), 41.4 (C-1′), 126.0 (C-3), 128.1 (C-4), 129.0 (C-2), 131.8 (C-1); m/z (C.I.) 154 (MH^+, 100%), 136 (45), 123 (16), 108 (14), 91 (40).

1-(Phenyloxycarbonyl)-1,2-dihydropyridine (**131**)[73]

Phenylchloroformate (18.75 cm^3, 150 mmol) was added dropwise to a stirred suspension of sodium borohydride (6.24 g) in pyridine (12.13 cm^3, 150 mmol) and methanol (120 cm^3) at -78°C. The rate of addition was controlled so as to maintain the temperature below -60°C. After 2 hours the reaction mixture was poured into ice-water (*ca.* 400 cm^3), allowed to warm to 20°C and extracted with ether (300, 2 x 200 cm^3). The combined extracts were dried ($MgSO_4$) and concentrated under reduced pressure. Recrystallisation from ethanol yielded the title compound as colourless needles (17.9 g, 59%): m.p. 65-66°C (from ethanol) (lit.[73] 65-67°C); R_F 0.60 (petrol-ethyl acetate 4:1); (Found: C, 71.9; H, 5.5; N, 6.9. Calc. for $C_{12}H_{11}NO_2$ C, 71.6; H, 5.5; N, 7.0%); ν_{max}(nujol) 1705 (C=O), 1380, 1355, 1325, 1200, 1065, 740, 700 cm^{-1}; m/z (E.I.) 201 (M^+, 46%), 200 (49), 156 (12), 124 (27), 108 (22), 94 (7), 80 (100).

1-(t-Butoxycarbonyl)-1,2-dihydropyridine (**132**)[73]

Potassium *tert*-butoxide (11.3 g, 101 mmol) in THF (170 cm^3) was added dropwise to a stirred solution of the dihydropyridine (**131**) (18.4 g, 91.5 mmol) in THF (200 cm^3) at 20°C under a nitrogen atmosphere. After 1 h the reaction mixture was poured into water (500 cm^3) and extracted with diethyl ether (3 x 300 cm^3). The combined extracts were washed with aqueous sodium hydroxide solution (0.5 M, 2 x 200 cm^3), dried ($MgSO_4$) and concentrated under reduced pressure to yield the title compound as a yellow oil (15.0 g, 90%). TLC indicated the title compound R_F 0.70 (petrol-ethyl acetate 4:1) plus a minor amount of the 1,4-isomer R_F 0.75. The product was used immediately as it tended to decompose on standing: δ_H(60 MHz, $CDCl_3$) 1.49 (9H, s, C*Me*$_3$), 4.32 (2H, dd, *J* 4, 2 Hz, 2 x 2-H), 5.10-5.85 (3H, m, 3-H, 4-H, 5-H), 6.63 (1H, br d, *J* 7 Hz, 6-H); m/z (E.I) 181 (M^+, 42%), 125 (100).

2-(t-Butoxycarbonyl)-7-(methoxycarbonyl)-2-azabicyclo[2.2.2]oct-5-ene **(133/134)**

A solution of the dihydropyridine (**132**) (15.0 g, 82.8 mmol) and methyl acrylate (14.9 cm^3, 165 mmol) in toluene (40 cm^3) was heated to reflux under a nitrogen atmosphere. After 2 days the solvent and excess methyl acrylate was evaporated under reduced pressure to leave an oil-solid mixture. Repeated recrystallisation from toluene yielded the *endo* isomer (**133**) as a colourless crystalline solid (6.58 g, 30%). Column chromatography (petrol-ethyl acetate 9:1) of the remaining material yielded the *exo* isomer (**134**) as a colourless oil (6.52 g, 29%).

endo isomer (**133**): m.p. 113-114°C (from toluene); R_F 0.50 (petrol-ethyl acetate 4:1); (Found: C, 63.0; H, 8.0; N, 5.3. $C_{14}H_{21}NO_4$ requires C, 62.9; H, 7.9; N, 5.2%); $\nu_{max}(CHCl_3)$ 1725 (C=O), 1665 (C=C), 1395, 1370, 1350, 1300, 1280, 1160, 1115 cm^{-1}; $\delta_H(CDCl_3)$ 1.46 (9H, s, C*Me*$_3$), 1.85 (2H, m, 2 x 8-H), 2.80 (1H, m, 4-H), 2.90 (1H, d, J_{gem} 10.3 Hz, 3-H), 3.06 (1H, m, 7-H), 3.22 (1H, dd, J_{gem} 10.3, $J_{3,4}$ 2.2 Hz, 3-H), 3.65 (3H, s, OMe), 5.06 (1H, br s, 1-H), 6.34 (1H, m, 5-H), 6.43 (1H, m, 6-H); m/z (C.I., NH_3) 285 (MNH_4^+, 2%), 268 (MH^+, 3), 229 (26), 212 (30), 168 (51), 81 (100).

exo isomer (**134**): R_F 0.50 (petrol-ethyl acetate 4:1); $\nu_{max}(CHCl_3)$ 1725 (C=O), 1665 (C=C), 1390, 1345, 1305, 1160, 1115 cm^{-1}; $\delta_H(CDCl_3)$ 1.43 (9H, 2 x s, C*Me*$_3$), 1.50 (1H, m, 8-H), 2.10 (1H, m, 8-H), 2.54 (1H, m, 7-H), 2.77 (1H, m, 4-H), 2.93 (1H, m, 3-H), 3.34 (1H, m, 3-H), 3.70 (3H, 2 x s, OMe), 4.94 (1H, m, 1-H), 6.44 (2H, m, 5-H, 6-H); m/z (C.I.) 268 (MH^+, 4%), 252 (4), 212 (100), 181 (24).

Methyl 5-[N-(t-butoxycarbonyl)aminomethyl]cyclohexa-1,3-diene-1-carboxylate **(135)**

A solution of 1,1,1,3,3,3-hexamethyldisilazane (7.30 cm^3, 34.6 mmol) in dry THF (35 cm^3) at -78°C was treated with a solution of *n*-butyl lithium in hexanes (1.6 M,

34.6 mmol) under a nitrogen atmosphere. After 20 min a solution of the adducts (**133** + **134**) (8.40 g, 31.5 mmol) in THF (90 cm^3) was added dropwise *via* cannular. The reaction mixture was stirred for 10 min at this temperature, allowed to warm to 20°C and quenched with saturated aqueous ammonium chloride solution (100 cm^3). The mixture was extracted with dichloromethane (600, 2 x 200 cm^3). The combined extracts were dried ($MgSO_4$) and concentrated under reduced pressure. Column chromatography (petrol-ethyl acetate 4:1) yielded the title compound as a colourless solid (7.00 g, 83%): m.p. 69-70°C (from petrol-ethyl acetate); R_F 0.30 (petrol-ethyl acetate 4:1); (Found: C, 62.8; H, 8.0; N, 5.1. $C_{14}H_{21}NO_4$ requires C, 62.9; H, 7.9; N, 5.2%); $\nu_{max}(CHCl_3)$ 3450 (NH), 1695 (C=O), 1640 (C=C), 1420, 1360, 1230, 1150, 1080 cm^{-1}; $\delta_H(CDCl_3)$ 1.44 (9H, s, CMe_3), 2.34 (1H, ddd, J_{gem} 19.5, $J_{6\beta,5}$ 13.5, $J_{6\beta,2}$ 1.5 Hz, 6β-H), 2.58 (1H, ddd partially obscured by 5-H, J_{gem} 19.5, $J_{6\alpha,5}$ 8.5, $J_{6\alpha,2}$ 1.5 Hz, 6α-H), 2.60 (1H, m, 5-H), 3.06 (1H, m, 1′-H), 3.23 (1H, m, 1′-H), 3.76 (3H, s, OMe), 4.72 (1H, br s , N-H), 6.03 (1H, dd, $J_{4,3}$ 9.5, $J_{4,5}$ 3.0 Hz, 4-H), 6.12 (1H, ddd, $J_{3,4}$ 9.5, $J_{3,2}$ 5.0, $J_{3,5}$ 1.5 Hz, 3-H), 6.99 (1H, dt, $J_{2,3}$ 5.0, $J_{2,6\alpha}$ 1.5, $J_{2,6\beta}$ 1.5 Hz, 2-H); $\delta_C(CDCl_3)$ 14.1 (CMe_3), 24.7 (C-6), 28.3 (CMe_3), 34.3 (C-5), 43.2 (C-1′), 51.6 (OMe), 124.6 (C-3), 126.5 (C-1), 132.6 (C-4), 134.8 (C-2), 155.9 (C=O), 167.5 (C=O); m/z (C.I) 268 (MH^+, 2%), 233 (3), 212 (41), 180 (18), 168 (100), 137 (70).

Methyl 5-(aminomethyl)cyclohexa-1,3-diene-1-carboxylate **(136)**

The BOC protected amine (**135**) (28.8 g, 0.11 mol) was added portion-wise to trifluoroacetic acid (100 cm^3). After stirring for 5 mins, excess TFA was evaporated under reduced pressure to leave a yellow oil. Sodium hydroxide (1.0 M, *ca.* 400 cm^3) was added cautiously until the solution was alkaline to litmus. The solution was extracted with ethyl acetate (2 x 500 cm^3), dried ($MgSO_4$) and evaporated to give the title compound as a yellow oil (15.7 g, 85%): R_F 0.80 (dichloromethane - methanol - aqueous ammonia 20:8:1); ν_{max}(liquid film) 1700 (C=O), 1630 (C=C),

1565, 1430, 1265, 1200, 1090, 715 cm^{-1}; $\delta_H(CDCl_3)$ 2.35 (1H, m, 6β-H), 2.51 (1H, m, 5-H), 2.60 (1H, m, 6α-H), 2.77 (2H, m, 2 x 1′-H), 3.73 (3H, s, OMe), 4.29 (2H, br s, NH_2), 6.02 (1H, dd, $J_{4,3}$ 9.5, $J_{4,5}$ 3.0 Hz, 4-H), 6.11 (1H, dd, $J_{3,4}$ 9.5, $J_{3,2}$ 5.3 Hz, 3-H), 6.96 (1H, d, $J_{2,3}$ 5.3 Hz, 2-H); $\delta_C(CDCl_3)$ 24.8 (C-6), 37.1 (C-5), 45.1 (C-1′), 51.6 (OMe), 124.3 (C-3), 126.1 (C-1), 132.8 (C-4), 135.8 (C-2), 167.8 (C=O); m/z (C.I) 168 (MH^+,100%), 151 (22), 137 (38), 105 (12), 91 (20).

Methyl 5-[N-(p-nitrobenzenesulphonyl)aminomethyl]cyclohexa-1,3-diene-1-carboxylate **(139)**

A solution of the amine (**136**) (15.7 g, 93.9 mmol) in dry THF (250 cm^3) was treated with *p*-nitrobenzenesulphonyl chloride (22.9 g, 0.103 mol) and triethylamine (14.4 cm^3, 0.103 mol). After stirring for 40 h at 20°C the reaction mixture was poured into water (500 cm^3) and extracted with ethyl acetate (3 x 500 cm^3). The combined extracts were dried ($MgSO_4$) and evaporated under reduced pressure to give a dark brown oil. Column chromatography (petrol-ethyl acetate 4:1 to 1:1) yielded the title compound as a pale yellow solid (20.7 g, 63%): m.p. 130-132°C (from petrol-ethyl acetate); R_F 0.30 (petrol-ethyl acetate 7:3); (Found: C, 51.1; H, 4.6; N, 7.9. $C_{15}H_{16}N_2O_6S$ requires C, 51.1; H, 4.6; N, 7.95%); $\nu_{max}(CHCl_3)$ 1695 (C=O), 1350 (SO_2), 1300, 1260, 1160 (SO_2), 1085 cm^{-1}; $\delta_H(D_6$-acetone) 2.31 (1H, ddd, J_{gem} 16.9, $J_{6\beta,5}$ 10.9, $J_{6\beta,2}$ 1.7 Hz, 6β-H), 2.53 (1H, ddd, J_{gem} 16.9, $J_{6\alpha,5}$ 8.4, $J_{6\alpha,2}$ 1.5 Hz, 6α-H), 2.62 (1H, m, 5-H), 3.03 (2H, m, 1′-H), 3.70 (3H, s, OMe), 6.06 (1H, ddd, $J_{4,3}$ 9.5, $J_{4,5}$ 3.5, $J_{4,2}$ 1.5 Hz, 4-H), 6.12 (1H, ddd, $J_{3,4}$ 9.5, $J_{3,2}$ 5.1, $J_{3,5}$ 1.5 Hz, 3-H), 6.93 (1H, m, 2-H), 7.01 (1H, br t, *J* 6.2 Hz, N-H), 8.12 (2H, d, *J* 9.0 Hz, Ar-H), 8.42 (2H, d, *J* 9.0 Hz, Ar-H); $\delta_C(CDCl_3)$ 24.5 (C-6), 33.8 (C-5), 45.6 (C-1′), 51.8 (OMe), 124.4 and 128.2 (aromatic CH), 125.4 (C-3), 126.2 (C-1), 132.5 (C-4), 133.1 (C-2), 145.8 and 150.0 (aromatic C), 167.5 (C=O); m/z (C.I) 353 (MH^+,9%), 321 (100), 291 (20), 156 (24).

Methyl 5-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]cyclohexa-1,3-diene-1-carboxylate (**140**)

Sodium hydride (0.28 g, 11.6 mmol) was added slowly to a stirred solution of the sulphonamide (**139**) (3.15 g, 8.94 mmol) in dry DMF (50 cm^3) over 7 min. The resultant black solution was stirred at 20°C for 1 h and then *p*-nitrobenzene-sulphonyl chloride (2.57 g, 11.6 mmol) added. After 1.5 h water (200 cm^3) was added and the mixture extracted with ethyl acetate (3 x 200 cm^3). The combined extracts were washed with brine (100 cm^3), dried ($MgSO_4$) and evaporated under reduced pressure to leave a dark brown oil. Column chromatography petrol-ethyl acetate 4:1 to 1:1) yielded the title compound as a pale yellow solid (3.37 g, 70%): m.p. 178-179°C (from petrol-ethyl acetate); R_F 0.50 (petrol-ethyl acetate 7:3); (Found: C, 46.9; H, 3.4; N, 7.6. $C_{21}H_{19}N_3O_{10}S_2$ requires C, 46.9; H, 3.6; N, 7.8%); $\nu_{max}(CHCl_3)$ 1700 (C=O), 1350 (SO_2), 1310, 1270, 1170 (SO_2), 1085 cm^{-1}; $\delta_H(CDCl_3)$ 2.39 (2H, m, 6-H), 2.92 (1H, m, 5-H), 3.73 (2H, m partially obscured by OMe, 2 x 1′-H), 3.75 (3H, s, OMe), 5.88 (1H, dd, $J_{4,3}$ 9.3, $J_{4,5}$ 4.4 Hz, 4-H), 6.16 (1H, ddd, $J_{3,4}$ 9.3, $J_{3,2}$ 5.2, $J_{3,5}$ 1.6 Hz, 3-H), 7.01 (1H, d, $J_{2,3}$ 5.2 Hz, 2-H), 8.28 (4H, d, *J* 9.0 Hz, Ar-H), 8.45 (4H, d, *J* 9.0 Hz, Ar-H); $\delta_C(CDCl_3)$ 24.3 (C-6), 32.7 (C-5), 51.0 (C-1′), 51.8 (OMe), 124.4 and 129.9 (aromatic CH), 126.0 (C-3), 126.6 (C-1), 131.4 (C-4), 132.1 (C-2), 144.5 and 150.9 (aromatic C), 166.7 (C=O); m/z (C.I., NH_3) 555 (MNH_4^+, 100%), 538 (MH^+, 18), 523 (15), 493 (31).

Reaction of diene disulphonimide (140) with potassium iodide

The diene disulphonimide (**140**) (195 mg, 0.36 mmol) and potassium iodide (132 mg, 0.80 mmol) were suspended in DMF (2 cm^3) and heated at 110°C under a nitrogen atmosphere for 16 h. TLC on silica gel indicated one major product plus a trace of unreacted starting material. The reaction mixture was diluted with water (35

cm^3) and extracted with ether (3 x 30 cm^3). The combined extracts were dried ($MgSO_4$) and evaporated to leave a brown oil. Column chromatography (petrol-ethyl acetate 9:1 to 17:3) gave recovered starting material (3.5 mg, 2%) and methyl 3-methylbenzoate (**142**) (35 mg, 64%) as a colourless oil: δ_H(60 MHz, $CDCl_3$) 2.50 (3H, s, Me), 3.98 (3H, s, OMe), 7.20-8.00 (4H, m, Ar-H); m/z (C.I.) 151 (MH^+, 100%), 119 (54), 91 (34).

Osmium tetroxide catalysed *cis*-hydroxylation of diene disulphonimide (140)

N-methylmorpholine *N*-oxide (0.62 g, 5.29 mmol) in acetone (10 cm^3) and water (25 cm^3) was treated with a 0.5% (w/v) solution of osmium tetoxide in *tert*-butanol (11.1 cm^3, 0.05 equiv). The diene disulphonimide (**140**) (2.37 g, 4.41 mmol) in THF (50 cm^3) was added slowly and the reaction mixture stirred at 20°C for 24 h. TLC (petrol-ethyl acetate 1:1) indicated three close running products and baseline material. THF and acetone were evaporated under reduced pressure and the reaction mixture diluted with water (400 cm^3), acidified with 2M hydrochloric acid and carefully extracted with ethyl acetate (1000 cm^3, 2 x 500 cm^3). The combined extracts were dried ($MgSO_4$) and preabsorbed onto silica gel. Column chromatography (petrol-ethyl acetate 54:46) yielded:

Methyl 1,2-dihydroxy-5β-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]cyclohex-3-ene-1-carboxylate (**145**) (0.25 g, 10%): m.p. 217-218°C (from ethyl acetate); R_F 0.32 (petrol-ethyl acetate 1:1); (Found: C, 44.0; H, 3.7; N, 7.2. $C_{21}H_{21}N_3O_{12}S_2$ requires C, 44.1; H, 3.7; N, 7.35%); ν_{max}(nujol) 1735 (C=O), 1540, 1360 (SO_2), 1310, 1170 (SO_2), 800 cm^{-1}; δ_H(D_6-DMSO) 1.52-1.72 (2H, m, 2 x 6-H), 2.84 (1H, m, 5-H), 3.65 (3H, s, OMe), 3.74 (1H, m, 1′-H), 4.00 (1H, m, 1′-H), 4.30 (1H, m, 2-H), 4.91 (1H, s, 1-OH), 5.03 (1H, d, $J_{2,OH}$ 7.9 Hz, 2-OH), 5.51 (2H, m, 3-H, 4-H), 8.21 (4H, d, *J* 9.2 Hz, Ar-H), 8.47 (4H, d, *J* 9.2 Hz, Ar-H); δ_C(D_6-DMSO) 32.4 (C-5), 34.9 (C-6), 52.1 (OMe), 53.8 (C-1′), 69.1 (C-2), 74.8 (C-1), 124.9 and 129.8

(aromatic CH), 126.8 (C-4), 130.9 (C-3), 143.5 and 150.9 (aromatic C), 175.2 (C=O); m/z (C.I) 554 (MH^+-H_2O, 4%), 536 (MH^+-$2H_2O$, 46%), 504 (100), 402 (52), 349 (60), 319 (70).

Methyl 3β,4β-dihydroxy-5β-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]cyclo-hex-1-ene-1-carboxylate (**144**) (0.20 g, 8%): m.p. 212-213°C (from ethyl acetate); R_F 0.25 (petrol-ethyl acetate 1:1); (Found: C, 43.7; H, 3.7; N, 7.2. $C_{21}H_{21}N_3O_{12}S_2$ requires C, 44.1; H, 3.7; N, 7.35%); ν_{max}(nujol) 3420 (OH), 1685 (C=O), 1350 (SO_2), 1310, 1175 (SO_2), 1145, 1090, 860, 810, 750 cm^{-1}; δ_H(D_6-DMSO) 1.90 (2H, m, 6-H), 2.23 (1H, m, 5-H), 3.63 (3H, s, OMe), 3.65 (1H, m partially obscured by OMe, 4-H), 3.98 (2H, m, 1′-H), 4.16 (1H, m, 3-H), 4.73 (1H, d, $J_{4,OH}$ 3.3 Hz, 4-OH), 5.17 (1H, d, $J_{3,OH}$ 6.4 Hz, 3-OH), 6.53 (1H, s, 2-H), 8.26 (4H, d, *J* 8.9 Hz, Ar-H), 8.49 (4H, d, *J* 8.9 Hz, Ar-H); δ_C(D_6-DMSO) 23.4 (C-6), 37.1 (C-5), 51.6 (OMe), 52.1 (C-1′), 66.3 (C-4), 68.5 (C-3), 124.9 and 129.8 (aromatic CH), 128.4 (C-1), 140.7 (C-2), 143.5 and 150.9 (aromatic C), 166.2 (C=O); m/z (C.I) 536 (MH^+-$2H_2O$, 11%), 504 (22), 388 (100).

Methyl 3α,4α-dihydroxy-5β-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]cyclo-hex-1-ene-1-carboxylate (**143**) (0.40g, 16%): m.p. 198-199°C (from petrol-ethyl acetate); R_F 0.21 (petrol-ethyl acetate 1:1); (Found: C, 44.0; H, 3.7; N, 7.3. $C_{21}H_{21}N_3O_{12}S_2$ requires C, 44.1; H, 3.7; N, 7.35%); ν_{max}(nujol) 3480 (OH), 3360 (OH), 1715 (C=O), 1520, 1350 (SO_2), 1245, 1165 (SO_2), 795, 740 cm^{-1}; δ_H(D_6-DMSO) 1.88 (1H, dd, J_{gem} 17.8, $J_{6\beta,5}$ 9.0 Hz, 6β-H), 2.18 (1H, dd, J_{gem} 17.8, $J_{6\alpha,5}$ 4.8 Hz, 6α-H), 2.37 (1H, m, 5-H), 3.43 (1H, m, 4-H), 3.62 (3H, s, OMe), 3.85 (1H, m, 1′-H), 4.10 (1H, m, 3-H), 4.14 (1H, m, 1′-H), 4.69 (1H, d, $J_{4,OH}$ 6.8 Hz, 4-OH), 5.11 (1H, d, $J_{3,OH}$ 6.2 Hz, 3-OH), 6.70 (1H, m, 2-H), 8.23 (4H, d, *J* 9.0 Hz, Ar-H), 8.48 (4H, d, *J* 9.0 Hz, Ar-H); δ_C(D_6-DMSO) 26.8 (C-6), 34.6 (C-5), 51.7 (OMe), 52.5 (C-1′), 64.7 (C-4), 70.2 (C-3), 124.9 and 129.8 (aromatic CH), 129.4 (C-1), 138.4 (C-2), 143.5 and 150.9 (aromatic C), 166.5 (C=O); m/z (C.I) 536 (MH^+-$2H_2O$, 6%), 504 (12), 388 (100).

'Wet' Prévost reaction upon diene disulphonimide (140)

A mixture of silver acetate (0.31 g, 1.86 mmol) and iodine (0.24 g, 0.93 mmol) in glacial acetic acid (20 cm^3) was stirred at 20°C until all the iodine was consumed. The diene disulphonimide (**140**) (0.50 g, 0.93 mmol) was added and the reaction mixture heated to reflux under a nitrogen atmosphere for 2 h. Water (0.2 cm^3) was added and heating continued for a further 2 h. The reaction mixture was allowed to cool, filtered and concentrated under reduced pressure (azeotroping with toluene). TLC indicated four products which were separated by column chromatography (petrol-ethyl acetate 7:3 then 1:1) to yield:

Methyl 3α,4α-diacetoxy-5β-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]cyclohex-1-ene-1-carboxylate (**153**) and *methyl 3β,4α-diacetoxy-5β-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]cyclohex-1-ene-1-carboxylate* (**154**) as an inseparable mixture, pale yellow solid (124 mg, 20%): R_F 0.78 (petrol - ethyl acetate 1:1).

Diacetate (**153**): $\delta_H(CDCl_3)$ 1.99 (3H, s, Me), 2.10 (1H, m, 6β-H), 2.12 (3H, s, Me), 2.53 (1H, dd, J_{gem} 18.4, $J_{6\alpha,5}$ 5.4 Hz, 6α-H), 2.73 (1H, m, 5-H), 3.75 (3H, s, OMe), 3.76 (1H, dd, J_{gem} 15.0, $J_{1',5}$ 8.2 Hz, 1'-H), 3.89 (1H, dd, J_{gem} 15.0, $J_{1',5}$ 6.0 Hz, 1'-H), 4.97 (1H, dd, $J_{4,5}$ 10.1, $J_{4,3}$ 4.0 Hz, 4-H), 5.67 (1H, dd, $J_{3,2}$ 5.0, $J_{3,4}$ 4.0 Hz, 3-H), 6.74 (1H, d m, $J_{2,3}$ 5.0 Hz, 2-H), 8.32 (4H, d, *J* 9.0 Hz, Ar-H), 8.45 (4H, d, *J* 9.0 Hz, Ar-H);

Diacetate (**154**): $\delta_H(CDCl_3)$ 2.05 (3H, s, Me), 2.06 (3H, s, Me), 2.10-2.60 (3H, m, 5-H, 2 x 6-H), 3.73 (3H, s, OMe), 3.70-3.83 (2H, m, 2 x 1'-H), 5.04 (1H, dd, $J_{4,5}$ 10.2, $J_{4,3}$ 7.1 Hz, 4-H), 5.56 (1H, d m, $J_{3,4}$ 7.1 Hz, 3-H), 6.62 (1H, t, $J_{2,3}$ 2.0, $J_{2,6}$ 2.0 Hz, 2-H), 8.30 (4H, d, *J* 9.0 Hz, Ar-H), 8.45 (4H, d, *J* 9.0 Hz, Ar-H).

Methyl 3β-acetoxy-4β-hydroxy-5β-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]-cyclohex-1-ene-1-carboxylate (**157**) as a pale yellow solid (19 mg, 3%): R_F 0.59 (petrol-ethyl acetate 1:1); $\delta_H(D_6DMSO)$ 2.00 (2H, m, 2 x 6-H), 2.06 (3H, s, Me),

2.35 (1H, m, 5-H), 3.65 (3H, s, OMe), 3.81 (1H, m, 4-H), 3.90 (1H, dd, J_{gem} 15.0, $J_{1',5}$ 7.5 Hz, 1'-H), 4.09 (1H, dd, J_{gem} 15.0, $J_{1',5}$ 6.8 Hz, 1'-H), 5.24 (2H, br s, 4-OH, 3-H), 6.45 (1H, br s, 2-H), 8.29 (4H, d, *J* 9.0 Hz, Ar-H), 8.49 (4H, d, *J* 9.0 Hz, Ar-H).

Methyl 3α-acetoxy-4α-hydroxy-5β-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]-cyclohex-1-ene-1-carboxylate (**155**) and *methyl 4α-acetoxy-3α-hydroxy-5β-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]cyclohex-1-ene-1-carboxylate* (**156**) as an inseparable mixture, pale yellow solid (294 mg, 52%): R_F 0.51 (petrol-ethyl acetate 1:1); m/z (E.I.) 613 (M^+, 3%), 571 (2), 553 (3), 367 (4), 317 (12), 186 (32), 153 (45), 122 (35), 43 (100).

Hydroxy acetate (**155**): $\delta_H(CDCl_3)$ 1.95-2.21 (1H, m, 6β-H), 2.16 (3H, s, Me), 2.40-2.59 (2H, m, 5-H, 6α-H), 3.70-3.92 (2H, m, 2 x 1'-H), 3.73 (3H, s, OMe), 4.18 (1H, dd, $J_{4,5}$ 11.0, $J_{4,3}$ 4.4 Hz, 4-H), 5.45 (1H, dd, $J_{3,2}$ 4.6, $J_{3,4}$ 4.4 Hz, 3-H), 6.79 (1H, dd, $J_{2,3}$ 4.6, $J_{2,6\beta}$ 1.6 Hz, 2-H), 8.33 (4H, d, *J* 9.2 Hz, Ar-H), 8.45 (4H, d, *J* 9.2 Hz, Ar-H).

Hydroxy acetate (**156**): $\delta_H(CDCl_3)$ 1.95-2.21 (1H, m, 6β-H), 2.02 (3H, s, Me), 2.40-2.59 (1H, m, 6α-H), 2.82 (1H, m, 5-H), 3.70-3.92 (2H, m, 2 x 1'-H), 3.74 (3H, s, OMe), 4.53 (1H, dd, $J_{3,2}$ 4.5, $J_{3,4}$ 4.0 Hz, 3-H), 4.87 (1H, dd, $J_{4,5}$ 10.0, $J_{4,3}$ 4.0 Hz, 4-H), 6.84 (1H, d m, $J_{2,3}$ 4.5 Hz, 2-H), 8.32 (4H, d, *J* 9.2 Hz, Ar-H), 8.44 (4H, d, *J* 9.2 Hz, Ar-H).

3α,4α-dihydroxy-5β-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]cyclohex-1-ene-1-carboxylate (**143**) as a pale yellow solid (15 mg, 3%): R_F 0.21 (petrol-ethyl acetate 1:1), identical with the sample prepared via the osmium tetroxide route.

***Methyl 3α,4α-dihydroxy-5β-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]-cyclohex-1-ene-1-carboxylate* (143)**

A solution of the mixed hydroxy-acetates (**155** + **156**) (3.00 g, 4.89 mmol) in

methanol (140 cm^3) was treated with dilute aqueous ammonia solution and stirred at 20°C under a nitrogen atmosphere for 23 h. Methanol was evaporated under reduced pressure and the resultant slurry was lyophilised to leave a pale yellow solid. Recrystallisation from ethyl acetate yielded the title compound as a cream coloured solid (1.94 g, 69%), identical with the sample prepared *via* the osmium tetroxide route.

General procedure for acetylation of the diols

A solution of the diol in pyridine (4 cm^3) was treated with acetic anhydride (0.5 cm^3), DMAP (catalytic) and stirred at 20°C for 24 h. The reaction mixture was poured into chloroform (30 cm^3) and washed with dilute aqueous hydrochloric acid (4 x 25 cm^3) and water (25 cm^3). The organic phase was dried ($MgSO_4$) and concentrated under reduced pressure (azeotroping with toluene). Column chromatography (petrol-ethyl acetate 7:3) yielded the diacetate.

Methyl 3α,4α-diacetoxy-5β-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]-cyclo-hex-1-ene-1-carboxylate **(153)**

Acetylation of the diol (**143**) (64.5 mg, 0.11 mmol) using the general procedure described above gave the title compound as a pale yellow solid (60 mg, 81%): m.p. 214-215°C (dec.) (from petrol-ethyl acetate); R_F 0.78 (petrol-ethyl acetate 1:1); (Found C, 45.8; H, 3.8; N, 6.3. $C_{25}H_{25}N_3O_{14}S_2$ requires C, 45.8; H, 3.8; N, 6.4%); ν_{max}(nujol) 1730 (C=O), 1705 (C=O), 1525, 1375 (SO_2), 1340, 1255, 1240, 1170 (SO_2), 730 cm^{-1}; δ_H($CDCl_3$) 1.99 (3H, s, Me), 2.10 (1H, m, 6β-H), 2.12 (3H, s, Me), 2.53 (1H, dd, J_{gem} 18.4, $J_{6\alpha,5}$ 5.4 Hz, 6α-H), 2.73 (1H, m, 5-H), 3.75 (3H, s, OMe), 3.76 (1H, dd, J_{gem} 15.0, $J_{1',5}$ 8.2 Hz, 1′-H), 3.89 (1H, dd, J_{gem} 15.0, $J_{1',5}$ 6.0 Hz, 1′-H), 4.97 (1H, dd, $J_{4,5}$ 10.1, $J_{4,3}$ 4.0 Hz, 4-H), 5.67 (1H, dd, $J_{2,3}$ 5.0, $J_{3,4}$ 4.0 Hz,

3-H), 6.74 (1H, d m, $J_{2,3}$ 5.0 Hz, 2-H), 8.32 (4H, d, *J* 9.0 Hz, Ar-H), 8.45 (4H, d, *J* 9.0 Hz, Ar-H); $\delta_C(CDCl_3)$ 20.8 (2 x Me), 27.6 (C-6), 33.1 (C-5), 51.1 (C-1′), 52.3 (OMe), 64.8 (C-4), 70.5 (C-3), 124.4 and 130.1 (aromatic CH), 132.0 (C-2), 133.0 (C-1), 144.1 and 151.0 (aromatic C), 165.8 (C=O), 169.8 (C=O), 170.0 (C=O); m/z (C.I., NH_3) 673 (MNH_4^+, 81%), 488 (100), 428 (28).

Methyl 3β,4β-diacetoxy-5β-[N,N-di(p-nitrobenzenesulphonyl)aminomethyl]-cyclohex-1-ene-1-carboxylate **(158)**

Acetylation of the diol (**144**) (55.5 mg, 0.10 mmol) using the general procedure described above gave the title compound as a pale yellow solid (55 mg, 86%): m.p. 187-188°C (dec.) (from petrol-ethyl acetate); R_F 0.77 (petrol-ethyl acetate 1:1); (Found C, 46.0; H, 3.85; N, 6.3. $C_{25}H_{25}N_3O_{14}S_2$ requires C, 45.8; H, 3.8; N, 6.4%); ν_{max}(nujol) 1755 (C=O), 1700 (C=O), 1525, 1375 (SO_2), 1340, 1215, 1160 (SO_2), 730 cm^{-1}; $\delta_H(CDCl_3)$ 2.02 (3H, s, Me), 2.08 (3H, s, Me), 2.21 (1H, m, 6β-H), 2.39 (1H, br dd, J_{gem} 17.5, $J_{6\alpha,5}$ 5.5 Hz, 6α-H), 2.51 (1H, m, 5-H), 3.63 (1H, dd, J_{gem} 15.0, $J_{1',5}$ 6.1 Hz, 1′-H), 3.78 (3H, s, OMe), 3.93 (1H, dd, J_{gem} 15.0, $J_{1',5}$ 8.2 Hz, 1′-H), 5.23 (1H, m, 4-H), 5.33 (1H, m, 3-H), 6.60 (1H, br s, 2-H), 8.32 (4H, d, *J* 9.1 Hz, Ar-H), 8.48 (4H, d, *J* 9.1 Hz, Ar-H); $\delta_C(CDCl_3)$ 20.6 (Me), 20.7 (Me), 24.8 (C-6), 35.7 (C-5), 49.9 (C-1′), 52.1 (OMe), 65.3 (C-4), 68.8 (C-3), 124.6 and 130.0 (aromatic CH), 131.3 (C-2), 134.3 (C-1), 143.8 and 151.0 (aromatic C), 165.7 (C=O), 169.8 (C=O), 170.5 (C=O).

Methyl 3α,4α-isopropylidenedioxy-5β-[N,N-di(p-nitrobenzenesulphonyl)amino-methyl]cyclohex-1-ene-1-carboxylate **(164)**

The diol disulphonimide (**143**) (2.80 g, 4.90 mmol) was suspended in 2,2-dimethoxypropane (10 cm^3) and acetone (10 cm^3). A catalytic amount of *p*-TSA was

added and the reaction mixture stirred at 20°C under a nitrogen atmosphere for 22 h. The suspension cleared and then a fine white precipitate formed. Evaporation under reduced pressure and trituration with acetone yielded the title compound as a white powder (2.80 g, 94%): m.p. 220-220.5°C (from ethyl acetate); R_F 0.72 (petrol-ethyl acetate 1:1); (Found : C, 47.1; H, 4.2; N, 6.9. $C_{24}H_{25}N_3O_{12}S_2$ requires C, 47.1; H, 4.1; N, 6.9%); ν_{max}(nujol) 1715 (C=O), 1520, 1390, 1350 (SO_2), 1240, 1165 (SO_2), 800, 740 cm^{-1}; δ_H(D_6-DMSO) 1.30 (3H, s, Me), 1.38 (3H, s, Me), 1.85 (1H, dd, J_{gem} 16.5, $J_{6\beta,5}$ 9.0 Hz, 6β-H), 2.11 (1H, m, 5-H), 2.20 (1H, dd, J_{gem} 16.5, $J_{6\alpha,5}$ 3.7 Hz, 6α-H), 3.64 (3H, s, OMe), 4.02 (3H, m, 4-H, 2 x 1′-H), 4.68 (1H, m, 3-H), 6.79 (1H, m, 2-H), 8.21 (4H, d, *J* 9.0 Hz, Ar-H), 8.49 (4H, d, *J* 9.0 Hz, Ar-H); m/z (C.I., NH_3) 629 (MNH_4^+, 100%), 612 (MH^+, 5), 596 (16), 571 (15), 444 (50), 386 (35), 369 (10).

Reaction of acetonide disulphoimide (164) with potassium iodide

The acetonide disulphonimide (**164**) (309 mg, 0.51 mmol) and dry potassium iodide (419 mg, 2.53 mmol) were suspended in dry DMF (4 cm^3) and heated at 130°C under a nitrogen atmosphere for 21 h. The reaction mixture was poured into water (50 cm^3) and extracted with ethyl acetate (3 x 50 cm^3). The combined extracts were washed with aqueous sodium thiosulphate solution (2% w/v, 25 cm^3), dried ($MgSO_4$) and concentrated under reduced pressure. TLC on silica gel (petrol-ethyl acetate 4:1) indicated three products and baseline material. Column chromatography (petrol-ethyl acetate 17:3 to 4:1) gave:

Methyl 3α,4α-isopropylidenedioxy-5-methylenecyclohex-1-ene-1-carboxylate (**166**) as a colourless oil (12.5 mg, 11%): R_F 0.49 (petrol-ethyl acetate 4:1); ν_{max}($CHCl_3$) 1715 (C=O), 1660 (C=C), 1375, 1305, 1230, 1160, 1100, 1075, 1040, 910 cm^{-1}; δ_H($CDCl_3$) 1.38 (3H, s, Me), 1.41 (3H, s, Me), 3.04 (1H, d, J_{gem} 19.0 Hz, 6-H), 3.17 (1H, dm, J_{gem} 19.0 Hz, 6-H), 3.78 (3H, s, OMe), 4.63 (1H, d, $J_{4,3}$ 5.2 Hz, 4-H), 4.70 (1H, m, 3-H), 5.14 (1H, m, 1′-H), 5.26 (1H, m, 1′-H), 6.74 (1H, m, 2-H); δ_C($CDCl_3$)

26.6 (Me), 28.0 (Me), 28.6 (C-6), 52.0 (OMe), 74.3 (C-4), 76.9 (C-3), 109.6 (CMe_2), 115.7 (C-1′), 130.6 (C-1), 135.3 (C-2), 139.9 (C-5), 166.8 (C=O); m/z (E.I.) 209 (M^+-CH_3, 209.0799 $C_{11}H_{13}O_4$ requires 209.0812, 75%), 167 (22), 149 (100), 135 (22), 107 (25).

Methyl 3α,4α-isopropylidenedioxy-5β-(iodomethyl)cyclohex-1-ene-1-carboxylate (**165**) as colourless crystals (56.5 mg, 32%): m.p. 69-71°C (from light petrol); R_F 0.44 (petrol-ethyl acetate 4:1); (Found: C, 40.8; H, 4.9. $C_{12}H_{17}IO_4$ requires C, 40.9; H, 4.9%); $\nu_{max}(CHCl_3)$ 1710 (C=O), 1655 (C=C), 1430, 1370, 1220, 1160, 1095, 1045 cm^{-1}; $\delta_H(CDCl_3)$ 1.40 (3H, s, Me), 1.44 (3H, s, Me), 1.56 (1H, m, 5-H), 2.05 (1H, ddt, J_{gem} 17.7, $J_{6\beta,5}$ 10.3, $J_{6\beta,3}$ 2.2, $J_{6\beta,2}$ 2.2 Hz, 6β-H), 2.69 (1H, br dd, J_{gem} 17.7, $J_{6\alpha,5}$ 4.4 Hz, 6α-H), 3.34 (1H, dd, J_{gem} 10.1, $J_{1',5}$ 6.4 Hz, 1′-H), 3.47 (1H, dd, J_{gem} 10.1, $J_{1',5}$ 3.7 Hz, 1′-H), 3.78 (3H, s, OMe), 4.01 (1H, dd, $J_{4,5}$ 9.2, $J_{4,3}$ 6.2 Hz, 4-H), 4.63 (1H, ddd, $J_{3,4}$ 6.2, $J_{3,2}$ 3.9, $J_{3,6\beta}$ 2.2 Hz, 3-H), 6.94 (1H, ddd, $J_{2,3}$ 3.9, $J_{2,6\beta}$ 2.2, $J_{2,6\alpha}$ 0.7 Hz, 2-H); $\delta_C(CDCl_3)$ 10.2 (C-1′), 25.8 (Me), 28.2 (Me), 29.0 (C-6), 38.6 (C-5), 52.1 (OMe), 71.3 (C-4), 76.7 (C-3), 109.5 (CMe_2), 133.4 (C-1), 133.5 (C-2), 166.5 (C=O); m/z (E.I.) 352 (M^+, 2%), 337 (66), 149 (33).

Homonuclear decoupling experiment data for (**165**):

Signal Irradiated (Chemical shift, δ)	**Observed Resonance**						
	2-H	3-H	4-H	1′-H	1′-H	6α-H	6β-H
Original resonance	ddd	ddd	dd	dd	dd	br dd	dddd
2-H (6.94)		dd	dd	dd	dd	dd	ddd
3-H (4.63)	dd		d	dd	dd	dd	ddd
5-H (4.63)	ddd	ddd	d	d	d	br d	ddd

Methyl 3α,4α-isopropylidenedioxy-5β-(O-formylhydroxymethyl)cyclohex-1-ene-1-carboxylate (**167**) as a colourless oil (24 mg, 17%): R_F 0.25 (petrol-ethyl acetate

4:1); $\nu_{max}(CHCl_3)$ 1715-1725 (C=O), 1375, 1230, 1160, 1095, 1050 cm^{-1}; $\delta_H(CDCl_3)$ 1.40 (3H, s, Me), 1.44 (3H, s, Me), 2.05 (2H, m, 5-H, 6β-H), 2.68 (1H, br dd, J_{gem} 21.0, $J_{6\alpha,5}$ 9.0 Hz, 6α-H), 3.78 (3H, s, OMe), 4.10 (1H, dd, $J_{4,5}$ 8.5, $J_{4,3}$ 6.0 Hz, 4-H), 4.20 (1H, dd, J_{gem} 11.0, $J_{1',5}$ 5.5 Hz, 1'-H), 4.38 (1H, dd, J_{gem} 11.0, $J_{1',5}$ 3.5 Hz, 1'-H), 4.63 (1H, ddd, $J_{3,4}$ 6.0, $J_{3,2}$ 3.5, $J_{3,6\beta}$ 1.5 Hz, 3-H), 6.95 (1H, ddd, $J_{2,3}$ 3.5, $J_{2,6\beta}$ 2.0, $J_{2,6\alpha}$ 1.0 Hz, 2-H), 8.10 (1H, d, J 0.5 Hz, CHO); $\delta_C(CDCl_3)$ 25.0 (C-6), 25.8 (Me), 28.1 (Me), 36.7 (C-5), 52.1 (OMe), 63.9 (C-1'), 71.1 (C-4), 73.9 (C-3), 109.4 (CMe_2), 133.0 (C-1), 133.7 (C-2), 160.8 (CHO), 166.6 (C=O); m/z (E.I) 270 (M^+, 0.4%), 255 (39), 181 (17), 167 (16), 149(100), 135 (11).

Homonuclear decoupling experiment data for (**167**):

Signal Iradiated (Chemical shift, δ)	**Observed Resonance** 2-H	3-H	4-H	1'-H	1'-H	6α-H
Original resonance	ddd	ddd	dd	dd	dd	br dd
6α-H (2.68)	dd	ddd	dd	dd	dd	
6β-H & 5-H (4.28)	dd	dd	d	d	d	d 1.0 Hz

***Methyl 3α,4α-isopropylidenedioxy-5β-(iodomethyl)cyclohex-1-ene-1-carboxylate* (165)**

A suspension of 18-crown-6 (0.57 g, 2.16 mmol) and dry potassium iodide (0.65 g, 3.93 mmol) in toluene (130 cm^3) was stirred at 20°C for 10 mins. The disulphonimide (**164**) was added and the reaction mixture heated to reflux under a nitrogen atmosphere. After 3 days additional potassium iodide (0.50 g, 3.01 mmol) and 18-crown-6 (0.50 g, 1.89 mmol) were added. After a total of 7 days, toluene was evaporated under reduced pressure, ethyl acetate was added and the suspension was

filtered and concentrated under reduced pressure. Column chromatography (petrol-ethyl acetate 4:1) yielded the title compound as colourless crystals (435 mg, 63%), identical with the sample described above.

***Methyl 3α,4α-isopropylidenedioxy-5β-[N-acetyl-N-(p-nitrobenzenesulphonyl)-aminomethyl]cyclohex-1-ene-1-carboxylate* (170)**

A suspension of 18-crown-6 (96 mg, 0.36 mmol) and dry potassium acetate (64 mg, 0.65 mmol) in toluene (20 cm^3) was stirred at 20°C for 10 mins. The disulphonimide (**164**) was added and the reaction mixture heated to reflux. After 7 h additional potassium acetate (32 mg, 0.32 mmol) and 18-crown-6 (47 mg, 0.18 mmol) were added. After a total of 18 h toluene was evaporated under reduced pressure. Column chromatography (petrol-ethyl acetate 7:3 to 11:9) yielded the title compound as a pale yellow solid (92 mg, 60%): m.p. 159-160°C (from petrol-ethyl acetate); R_F 0.70 (petrol-ethyl acetate 1:1); (Found: C, 51.1; H, 5.1; N, 5.9. $C_{20}H_{24}N_2O_9S$ requires C, 51.3; H, 5.2; N, 6.0%); $\nu_{max}(CHCl_3)$ 1715 (C=O), 1370, 1350 (SO_2), 1220, 1170 (SO_2), 1090, 1055 cm^{-1}; $\delta_H(CDCl_3)$ 1.40 (3H, s, Me), 1.47 (3H, s, Me), 2.00 (1H, ddt, J_{gem} 17.5, $J_{6\beta,5}$ 9.9, $J_{6\beta,2}$ 2.0, $J_{6\beta,3}$ 2.0 Hz, 6β-H), 2.25 (1H, m, 5-H), 2.36 (3H, s, Me), 2.70 (1H, dd, J_{gem} 17.5, $J_{6\alpha,5}$ 4.3 Hz, 6α-H), 3.78 (3H, s, Me), 3.87 (1H, dd, J_{gem} 14.6, $J_{1',5}$ 8.6 Hz, 1′-H), 4.04 (1H, dd, $J_{4,5}$ 9.2, $J_{4,3}$ 6.2 Hz, 4-H) 4.09 (1H, dd, J_{gem} 14.6, $J_{1',5}$ 5.9 Hz, 1′-H), 4.67 (1H, m, 3-H), 6.97 (1H, m, 2-H), 8.10 (2H, d, *J* 8.8 Hz, Ar-H), 8.40 (2H, d, *J* 8.8 Hz, Ar-H); $\delta_C(CDCl_3)$ 24.9 (Me), 25.5 (C-6), 25.8 (Me), 27.9 (C-5), 37.8 (Me), 48.8 (C-1′), 52.1 (OMe), 71.2 (C-4), 75.6 (C-3), 109.5 (CMe_2), 124.4 and 129.0 (aromatic CH), 132.8 (C-1), 133.8 (C-2), 145.0 and 150.6 (aromatic C), 166.5 (C=O), 170.2 (C=O); m/z (C.I., NH_3) 486 (MNH_4^+, 22%), 453 (M^+-CH_3, 5), 444 (16), 439 (7), 411 (68), 381 (100).

Methyl 3α,4α-isopropylidenedioxy-5β-(acetoxymethyl)cycloclohex-1-ene-1-carboxylate (**169**)

(a) A solution of the iodide (**165**) (116 mg, 0.33 mmol) in dry DMF (5 cm^3) was treated with sodium acetate and heated to 110°C under an argon atmosphere for 2.5 h. The reaction mixture was poured into water (50 cm^3) and extracted with diethyl ether (4 x 25 cm^3). The combined extracts were dried ($MgSO_4$) and concentrated under reduced pressure. Column chromatography (petrol-ethyl acetate 4:1) yielded the title compound as a colourless oil (59 mg, 63%): R_F 0.80 (petrol-ethyl acetate 1:1); $\nu_{max}(CHCl_3)$ 1710 (C=O), 1645 (C=C), 1430, 1380, 1370, 1220, 1160, 1050 cm^{-1}; $\delta_H(CDCl_3)$ 1.36 (3H, s, Me), 1.40 (3H, s, Me), 2.02 (2H, m, 5-H, 6β-H), 2.03 (3H, s, Me), 2.61 (1H, dd, J_{gem} 21.0, $J_{6\alpha,5}$ 8.0 Hz, 6α-H), 3.74 (3H, s, OMe), 4.04 (1H, dd, J_{gem} 11.0, $J_{1',5}$ 6.0 Hz, 1'-H), 4.07 (1H, dd, $J_{4,5}$ 8.5, $J_{4,3}$ 6.0 Hz, 4-H), 4.24 (1H, dd, J_{gem} 11.0, $J_{1',5}$ 4.0 Hz, 1'-H), 4.58 (1H, m, 3-H), 6.89 (1H, m, 2-H); $\delta_C(CDCl_3)$ 20.8 (Me), 24.8 (C-6), 25.7 (Me), 28.0 (Me), 36.5 (C-5), 52.0 (OMe), 64.3 (C-1'), 71.0 (C-4), 73.9 (C-3), 109.2 (CMe_2), 132.8 (C-1), 133.7 (C-2), 166.6 (C=O), 170.8 (C=O); m/z (C.I.) 285 (MH^+, 1%), 269 (36), 227 (20), 195 (21), 167 (100), 149 (67).

(b) The diol (**190**) (80 mg, 0.33 mmol) and *p*-TSA (catalytic) were dissolved in acetone (3 cm^3) and 2,2-dimethoxypropane (3 cm^3). The reaction mixture was stirred at 20°C under a nitrogen atmosphere for 18 h after which solvents were evaporated under reduced pressure. Column chromatography (petrol-ethyl acetate 3:1) yielded the title compound as a colourless oil (88 mg, 94 %), identical with the sample described above.

Methyl 3α,4α-isopropylidenedioxy-5β-(hydroxymethyl)cyclohex-1-ene-1-carboxylate (**172**)

(a) A solution of the acetate (**169**) (85 mg, 0.30 mmol) in methanol (4 cm^3) was treated with aqueous ammonia solution (1 cm^3) and stirred at 20°C under a nitrogen atmosphere for 48 h. The solvents were evaporated under reduced pressure. Column chromatography (petrol - ethyl acetate 1:1) yielded the title compound as a colourless oil which crystallised on standing (63.5 mg, 87%): m.p. 52-53°C (from petrol-diethyl ether); R_F 0.40 (petrol-ethyl acetate 1:1); (Found: C, 59.3; H, 7.6. $C_{12}H_{18}O_5$ requires C, 59.5; H, 7.5%); $\nu_{max}(CHCl_3)$ 3500 (OH), 1710 (C=O), 1655 (C=C), 1435, 1375, 1250, 1160, 1100, 1040, 910 cm^{-1}; $\delta_H(CDCl_3)$ 1.40 (3H, s, Me), 1.47 (3H, s, Me), 1.96 (1H, m, 5-H), 1.98 (1H, m, 6β-H), 2.13 (1H, br s, OH), 2.60 (1H, dd, J_{gem} 22.0, $J_{6\alpha,5}$ 9.0 Hz, 6α-H), 3.75 (2H, m, 2 x 1′-H), 3.77 (3H, s, OMe), 4.16 (1H, dd, $J_{4,5}$ 8.5, $J_{4,3}$ 6.0 Hz, 4-H), 4.62 (1H, ddd, $J_{3,4}$ 6.0, $J_{3,2}$ 3.5, $J_{3,6\beta}$ 1.0 Hz, 3-H), 6.94 (1H, m, 2-H), $\delta_C(CDCl_3)$ 24.8 (C-6), 25.8 (Me), 28.0 (Me), 39.0 (C-5), 52.1 (OMe), 64.9 (C-1′), 71.4 (C-4), 76.4 (C-3), 109.4 (CMe_2), 133.5 (C-1,C-2), 166.7 (C=O); m/z (E.I.) 242 (M^+, 1%), 227 (65), 211 (4), 195 (3), 167 (22), 153 (45), 137 (100).

(b) The triol (**173**) (60 mg, 0.30 mmol) and *p*-TSA (catalytic) were dissolved in acetone (3 cm^3) and 2,2-dimethoxypropane (3 cm^3). The reaction mixture was stirred at 20°C under a nitrogen atmosphere for 22 h after which the solvents were evaporated under reduced pressure. Column chromatography (petrol-ethyl acetate 4:1 then 1:1) yielded the title compound as a colourless oil which crystallised on standing (60.5 mg, 84%), identical with the sample described above and the ketal (**191**) as a colourless oil (12.5 mg, 8%): R_F 0.80 (petrol-ethyl acetate 1:1); $\delta_H(CDCl_3)$ 1.33 (3H, s, Me), 1.39 (3H, s, Me), 1.42 (3H, s, Me), 1.95-2.20 (2H, m, 5-H, 6β-H), 2.64 (1H, m, 6α-H), 3.35-3.57 (2H, m, 1′-H), 3.76 (3H, s, OMe), 4.15 (1H, m, 4-H), 4.58 (1H, m, 3-H), 6.87 (1H, m, 2-H); $\delta_C(CDCl_3)$ 24.8 (Me), 26.0

(Me), 28.2 (Me), 37.3 (C-5), 52.0 (OMe), 60.5 and 60.7 (C-1′), 71.1 (C-4), 74.0 and 74.1 (C-3), 99.8 (CMe_2), 108.8 (CMe_2), 132.9 (C-1), 133.8 and 133.9 (C-2), 167.0 (C=O).

***Methyl 3α,4α-dihydroxy-5β-(hydroxymethyl)cyclohex-1-ene-1-carboxylate* (173)**

(a) The acetonide formate (**167**) (38 mg, 0.14 mmol) was dissolved in dry methanol (3 cm^3), Amberlyst-15 ion exchange resin was added and the reaction mixture stirred at 20°C for 17 h. The resin was filtered off and the methanol evaporated under reduced pressure. The residue was dissolved in glacial acetic acid (1 cm^3), water (1 cm^3) and THF (1 cm^3) and heated at 60°C under an argon atmosphere for 3.5 h. Removal of the solvents under reduced pressure and column chromatography (ethyl acetate) yielded the title compound as a white powder (18 mg, 64%): m.p. 115-115.5°C (from ethyl acetate); R_F 0.25 (ethyl acetate); (Found: C, 53.3; H, 7.1. $C_9H_{14}O_5$ requires C, 53.5; H, 7.0%); ν_{max}(nujol) 3250 br (OH), 1715 (C=O), 1655 (C=C), 1425, 1270, 1245, 1115, 1070, 1040 cm^{-1}; $\delta_H(D_2O)$ 2.11 (2H, m, 5-H, 6β-H), 2.55 (1H, m, 6α-H), 3.64 (1H, dd, J_{gem} 12.0, $J_{1',5}$ 6.0 Hz, 1′-H), 3.73 (2H, m, 4-H, 1′-H), 3.75 (3H, s, OMe), 4.28 (1H, dd, $J_{3,2}$ 4.5, $J_{3,4}$ 4.5 Hz, 3-H), 6.83 (1H, dd, $J_{2,3}$ 4.5, $J_{2,6\beta}$ 1.5 Hz, 2-H); $\delta_C(D_2O)$ 25.4 (C-6), 35.9 (C-5), 51.9 (OMe), 61.7 (C-1′), 64.8 (C-4), 68.7 (C-3), 131.4 (C-1), 135.9 (C-2), 168.9 (C=O); m/z (C.I., NH_3) 220 (MNH_4^+, 100%), 202 (M^+, 22), 185 (33), 170 (5), 153 (15).

(b) A solution of the acetonide (**172**) (40 mg, 0.17 mmol) in glacial acetic acid (1.5 cm^3), water (1.5 cm^3) and THF (1.5 cm^3) was heated to 50-60°C under an argon atmosphere for 17 h. Evaporation of the solvents under reduced pressure and column chromatography (ethyl acetate) yielded the title compound as a white powder (23.5 mg, 70%), identical with the sample described above.

(c) A solution of the acetate (**190**) (30 mg, 0.12 mmol) in methanol (3 cm^3) was treated with aqueous ammonia solution (1 cm^3) and stirred at 20°C under a nitrogen

atmosphere for 21 h. The solvents were evaporated under reduced pressure. Column chromatography (ethyl acetate) yielded the title compound as a white powder (21 mg, 84%), identical with the sample described above.

3α,4α-Dihydroxy-5β-(hydroxymethyl)cyclohex-1-ene-1-carboxylic acid (**174**)

A solution of the triol (**173**) (34 mg, 0.17 mmol) in water (3 cm^3) was treated with aqueous sodium hydroxide solution (1.0 M, 0.17 cm^3) and stirred at 20°C under an argon atmosphere for 5.5 h. The solution was acidified by the addition of IR-120 ion-exchange resin. The resin was filtered off and washed with water. The combined aqueous phases were lyophilised to yield the title compound as a white powder (32 mg, quantitative): R_F 0.83 (reverse phase silica, water); ν_{max}(nujol) 3410 (OH), 3370 (OH), 3280 (OH), 1685 (C=O), 1645 (C=C), 1410, 1295, 1265, 1120, 1095, 1075, 1025, 1005 cm^{-1}; $\delta_H(D_2O)$ 2.09 (2H, m, 5-H, 6β-H), 2.53 (1H, dd, J_{gem} 22.0, $J_{6\alpha,5}$ 9.0 Hz, 6α-H), 3.64 (1H, dd, J_{gem} 11.2, $J_{1',5}$ 6.0 Hz, 1′-H), 3.74 (2H, m, 4-H, 1′-H), 4.28 (1H, dd, $J_{3,4}$ 4.5, $J_{3,2}$ 4.5 Hz, 3-H), 6.79 (1H, dd, $J_{2,3}$ 4.5, $J_{2,6\beta}$ 2.0 Hz, 2-H); $\delta_C(D_2O)$, 25.8 (C-6), 36.2 (C-5), 62.0 (C-1′), 65.1 (C-4), 69.0 (C-3), 132.4 (C-1), 135.6 (C-2), 170.9 (C=O); m/z (C.I., NH_3) 206 (MNH_4^+, 100%), 190 (21), 188(12).

Homonuclear decoupling experiment data for (**174**):

Signal Irradiated	**Observed Resonance**				
(Chemical shift, δ)	2-H	3-H	4-H	1′-H	1′-H
Original resonance	dd	dd	m	m	dd
3-H (4.28)	d 2.0 Hz		d 9.0 Hz	dd 11.0 Hz 4.2 Hz	dd
4-H, 1′-H (3.74)	dd	d			m
6β-H, 5-H (2.09)	d 4.5 Hz	dd	m	d 11.0 Hz	d 11.0 Hz

Methyl 3α,4α-diacetoxy-5β-(acetoxymethyl)cyclohex-1-ene-1-carboxylate (**175**)

A solution of the triol (**173**) (12 mg, 0.06 mmol) in pyridine (1 cm^3) was treated with acetic anhydride (0.5 cm^3) and DMAP (catalytic) and stirred at 20°C for 15 h. The reaction mixture was poured into ethyl acetate (10 cm^3) and washed with dilute aqueous hydrochloric acid (3 x 10 cm^3) and water (10 cm^3). The organic phase was dried ($MgSO_4$) and concentrated under reduced pressure (azeotroping with toluene) to yield the title compound as a colourless oil (17 mg, 87%): R_F 0.74 (petrol-ethyl acetate 1:1); $\delta_H(CDCl_3)$ 2.04 (3H, s, Me), 2.06 (3H, s, Me), 2.09 (3H, s, Me), 2.33 (1H, dddd, J_{gem} 18.0, $J_{6\beta,5}$ 9.9, $J_{6\beta,2}$ 2.4, $J_{6\beta,3}$ 1.2 Hz, 6β-H), 2.49 (1H, m, 5-H), 2.75 (1H, br dd, J_{gem} 18.0, $J_{6\alpha,5}$ 5.2 Hz, 6α-H), 3.78 (3H, s, OMe), 4.09 (1H, dd, J_{gem} 11.2, $J_{1',5}$ 3.8 Hz, 1′-H), 4.20 (1H, dd, J_{gem} 11.2, $J_{1',5}$ 5.5 Hz, 1′-H), 5.04 (1H, dd, $J_{4,5}$ 10.8, $J_{4,3}$ 3.9 Hz, 4-H), 5.65 (1H, br dd, $J_{3,2}$ 5.3, $J_{3,4}$ 3.9 Hz, 3-H), 6.78 (1H, ddd, $J_{2,3}$ 5.3, $J_{2,6\beta}$ 2.4, $J_{2,6\alpha}$ 1.3 Hz, 2-H); $\delta_C(CDCl_3)$ 20.8 (3 x Me), 27.3 (C-6), 32.5 (C-5), 52.2 (OMe), 63.2 (C-1′), 64.9 (C-4), 69.0 (C-3), 131.8 (C-2), 134.3 (C-1), 166.3 (C=O), 170.1 (C=O), 170.9 (2 x C=O).

Methyl 3α,4α-dihydroxy-5β-(iodomethyl)cyclohex-1-ene-1-carboxylate (**176**)

A solution of the acetonide (**165**) (51 mg, 0.14 mmol) in THF (2.5 cm^3), glacial acetic acid (2 cm^3) and water (2 cm^3) was heated at 60°C under a nitrogen atmosphere for 39 h. The solvents were evaporated under reduced pressure. Column chromatography (petrol-ethyl acetate 1:1) yielded the title compound as a colourless gum (36 mg, 80%): R_F 0.36 (petrol-ethyl acetate 1:1); $\nu_{max}(CHCl_3)$ 3550 (OH), 3390 (OH), 1715 (C=O), 1655 (C=C), 1435, 1250, 1090, 1070, 910 cm^{-1}; $\delta_H(CDCl_3)$ 1.72 (1H, m, 5-H), 2.17 (1H, dddd, J_{gem} 18.3, $J_{6\beta,5}$ 10.3, $J_{6\beta,2}$ 2.6, $J_{6\beta,3}$ 1.4 Hz, 6β-H), 2.30 (2H, br s, 2 x OH), 2.68 (1H, br dd, J_{gem} 18.3, $J_{6\alpha,5}$ 5.1 Hz, 6α-H), 3.47 (2H, m, 2 x 1′-H), 3.61 (1H, dd, $J_{4,5}$ 10.3, $J_{4,3}$ 4.2 Hz, 4-H), 3.77 (3H, s, OMe), 4.33

(1H, m, 3-H), 6.88 (1H, ddd, $J_{2,3}$ 5.1, $J_{2,6\beta}$ 2.6, $J_{2,6\alpha}$ 1.1 Hz, 2-H); $\delta_C(CDCl_3)$ 11.7 (C-1′), 30.9 (C-6), 35.8 (C-5), 52.2 (OMe), 65.4 (C-4), 71.9 (C-3), 132.8 (C-1), 135.4 (C-2), 166.8 (C=O); m/z (C.I., NH_3) 330 (MNH_4^+, 100%), 312 (M^+, 10), 204 (59), 202 (51), 186 (18), 169 (15), 153 (8).

Homonuclear decoupling experiment data for (**176**):

Signal Irradiated (Chemical shift, δ)	Observed Resonance				
	2-H	3-H	4-H	6α-H	6β-H
Original resonance	ddd	m	dd	br dd	dddd
2-H (6.88)		br d	dd	dd	ddd
3-H (4.33)	br d		d	br dd	ddd
6α-H (2.68)	dd	br d	dd		ddd

3α,4α-dihydroxy-5β-(iodomethyl)cyclohex-1-ene-1-carboxylic acid (**177**)

(a) A solution of the methyl ester (**176**) (30 mg, 0.096 mmol) in THF (2 cm^3) was treated with aqueous sodium hydroxide solution (1.0 M, 0.10 cm^3). Water (*ca.* 1 cm^3) was added to give a homogeneous solution which was stirred at 20°C under a nitrogen atmosphere for 24 h. The solution was acidified by adding Amberlite IRA-120 (+) ion-exchange resin, filtered and the resin washed with water. The combined aqueous phases were lyophilsed to yield the title compound as an off-white flocculent solid (29 mg, quantitative): R_F 0.62 (ethyl acetate - acetic acid 99:1), $\delta_H(D_2O)$ 1.73 (1H, m, 5-H), 2.17 (1H, dddd, J_{gem} 18.0, $J_{6\beta,5}$ 10.0, $J_{6\beta,2}$ 2.0, $J_{6\beta,3}$ 1.2 Hz, 6β-H), 2.59 (1H, br dd, J_{gem} 18.0, $J_{6\alpha,5}$ 5.0 Hz, 6α-H), 3.43 (1H, dd, J_{gem} 10.2, $J_{1',5}$ 5.6 Hz, 1′-H), 3.48 (1H, dd, J_{gem} 10.2, $J_{1',5}$ 3.9 Hz, 1′-H), 3.64 (1H, dd, $J_{4,5}$ 10.1, $J_{4,3}$ 4.1 Hz, 4-H), 4.31 (1H, m, 3-H), 6.85 (1H, ddd, $J_{2,3}$ 4.5, $J_{2,6\beta}$ 2.0,

$J_{2,6\alpha}$ 0.8 Hz, 2-H); $\delta_C(D_2O)$ 11.7 (C-1′), 30.1 (C-6), 34.4 (C-5), 64.7 (C-4), 71.2 (C-3), 131.7 (C-1), 136.1 (C-2), 170.0 (C=O); m/z (C.I., NH_3) 316 (MNH_4^+, 25%), 298 (M^+, 1), 188 (100), 172 (20), 152 (20).

(b) The acetonide (**178**) (14 mg, 0.041 mmol) was treated with glacial acetic acid (1.2 cm^3) and water (1 cm^3). THF (1 cm^3) was added to give a homogeneous solution which was heated at 60°C under a nitrogen atmosphere for 24 h. The solvents were evaporated under reduced pressure and the residue dissolved in water (1 cm^3) and lyophilised to yield the title compound as a pale brown solid (12 mg, 98%), identical with the sample described above.

***3α,4α-Isopropylidenedioxy-5β-(iodomethyl)cyclohex-1-ene-1-carboxylic acid* (178)**

A solution of the methyl ester (**165**) (51 mg, 0.14 mmol) in THF (2.5 cm^3) was treated with aqueous sodium hydroxide solution (1.0 M, 0.15 cm^3). Water (*ca.* 1.2 cm^3) was added to give a homogeneous solution which was stirred at 20°C under a nitrogen atmosphere for 18 h. THF was evaporated under reduced pressure and the remaining aqueous solution diluted with water (15 cm^3) and washed with diethyl ether (5 cm^3). The aqueous solution was acidified by adding Amberlite IRA-120 (+) ion-exchange resin, filtered and lyophilised to yield the title compound as a cream solid (35 mg, 71%): R_F 0.82 (ethyl acetate-acetic acid 99:1); ν_{max}(nujol) 1680 (C=O), 1640 (C=C), 1290, 1250, 1215, 1065, 935 cm^{-1}; $\delta_H(CD_3OD)$ 1.47 (3H, s, Me), 1.50 (3H, s, Me), 1.62 (1H, m, 5-H), 2.09 (1H, ddt, J_{gem} 17.5, $J_{6\beta,5}$ 10.1, $J_{6\beta,2}$ 2.0, $J_{6\beta,3}$ 2.0 Hz, 6β-H), 2.75 (1H, dd, J_{gem} 17.5, $J_{6\alpha,5}$ 4.0 Hz, 6α-H), 3.41 (1H, dd, J_{gem} 10.0, $J_{1',5}$ 7.0 Hz, 1′-H), 3.61 (1H, dd, J_{gem} 10.0, $J_{1',5}$ 3.5 Hz, 1′-H), 4.11 (1H, dd, $J_{4,5}$ 9.0, $J_{4,3}$ 6.1 Hz, 4-H), 4.79 (1H, m, 3-H), 6.96 (1H, m, 2-H); $\delta_C(CD_3OD)$ 10.1 (C-1′), 26.2 (Me), 28.7 (Me), 30.1 (C-6), 40.9 (C-5), 73.0 (C-4), 78.3 (C-3), 110.8 (CMe_2), 134.8 (C-2); m/z (C.I., NH_3) 356 (MNH_4^+, 50%), 339 (MH^+, 50), 323

(100), 298 (20), 281 (15), 263 (19), 213 (35).

Osmium tetroxide catalysed *cis*-hydroxylation of the diene (135)

A solution of the diene (**135**) (7.00 g, 26.2 mmol) in acetone (100 cm^3) was treated with a solution of osmium tetroxide in *tert*-butanol (0.5% w/v, 1.0 cm^3), *N*-methylmorpholine *N*-oxide (3.37 g, 28.8 mmol) and water (1.0 cm^3). The reaction mixture was stirred at 20°C for 48 h and then concentrated under reduced pressure to yield a black oil-solid mixture. Trituration with diethyl ether and recrystallisation from ethyl acetate yielded *Methyl 3α,4α-dihydroxy-5β-[N-(t-butoxycarbonyl)-aminomethyl]cyclohex-1-ene-1-carboxylate* (**179**) as a colourless solid (2.25 g, 29%): m.p. 181-182°C (from ethyl acetate); R_F 0.28 (petrol-ethyl acetate 1:1); (Found: C, 56.0; H, 7.8; N, 4.65. $C_{14}H_{23}NO_6$ requires C, 55.8; H, 7.7; N, 4.65%); ν_{max}(nujol) 3400 (OH), 3280 (OH), 1705 (C=O), 1670 (C=C), 1530, 1285, 1255, 1165, 1100 cm^{-1}; δ_H(D_6-DMSO) 1.38 (9H, s, C*Me*$_3$), 1.86 (1H, br dd, J_{gem} 18.0, $J_{6\beta,5}$ 8.5 Hz, 6β-H), 1.96 (1H, m, 5-H), 2.38 (1H, br dd, J_{gem} 18.0, $J_{6\alpha,5}$ 4.0 Hz, 6α-H), 2.84 (1H, ddd, J_{gem} 14.0, $J_{1',5}$ 8.0, $J_{1',NH}$ 5.5 Hz, 1′-H), 3.12 (1H, dt, J_{gem} 14.0, $J_{1',5}$ 5.5, $J_{1',NH}$ 5.5 Hz, 1′-H), 3.39 (1H, m, 4-H), 4.06 (1H, m, 3-H), 4.52 (1H, d, $J_{OH,4}$ 5.5 Hz, 4-OH), 4.89 (1H, d, $J_{OH,3}$ 6.0 Hz, 3-OH), 6.66 (1H, br d, $J_{2,3}$ 4.0 Hz, 2-H), 6.80 (1H, br t, *J* 5.5 Hz, NH); δ_C(D_6-DMSO) 26.7 (C-6), 28.4 (C*Me*$_3$), 35.8 (C-5), 41.6 (C-1′), 51.8 (OMe), 65.0 (C-4), 69.6 (C-3), 77.7 (*C*Me$_3$), 130.0 (C-1), 138.6 (C-2), 156.1 (C=O), 166.9 (C=O); m/z (C.I.) 302 (MH^+, 1%), 268 (3), 246 (12), 228 (100), 184 (35), 152 (28).

The remaining material was purified by column chromatography (petrol-ethyl acetate 1:1) to yield *Methyl 3β,4β-dihydroxy-5β-[N-(t-butoxy-carbonyl)amino-methyl]cyclohex-1-ene-1-carboxylate* (**180**) as a colourless oil (3.08 g, 39%): R_F 0.28 (petrol-ethyl acetate 1:1); (Found: C, 56.2; H, 7.6; N, 4.7. $C_{14}H_{23}NO_6$ requires C, 55.8; H, 7.7; N, 4.65%); ν_{max}(liquid film) 3380 (OH), 1700 (C=O), 1520, 1435,

1365, 1270, 1250, 1165, 755 cm^{-1}; δ_H(D_6-DMSO) 1.38 (9H, s, C*Me*$_3$), 1.82 (2H, m, 5-H, 6-H), 2.15 (1H, d m, J_{gem} 16.0 Hz, 6α-H), 2.99 (2H, m, 2 x 1′-H), 3.67 (3H, s, OMe), 3.70 (3H, br s, 4-H), 4.12 (1H, br s, 3-H), 4.39 (1H, br s, 4-OH), 5.00 (1H, br s, 3-OH), 6.53 (1H, br s, 2-H), 6.83 (1H, br t, *J* 5.5 Hz, NH); δ_C(D_6-DMSO) 24.1 (C-6), 28.2 (C*Me*$_3$), 37.0 (C-5), 42.5 (C-1′), 51.5 (OMe), 66.9 (C-4), 68.8 (C-3), 77.5 (*C*Me$_3$), 129.1 (C-1), 140.8 (C-2), 155.8 (C=O), 166.5 (C=O); m/z (C.I.) 302 (MH^+, 28%), 284 (5), 268 (5), 246 (95), 228 (90), 202 (100), 184 (50).

Homonuclear decoupling experiment data for (**179**):

Signal Irradiated	**Observed Resonance**				
(Chemical shift, δ)	2-H	3-OH	4-H	1′-H	1′-H
Original resonance	br d	d	m	ddd	ddd
3-H (4.06)	br s	s	dd 5.5, 8.5 Hz	ddd	ddd
4-OH (4.52)	br d	d	dd 3.5, 8.5 Hz	ddd	ddd
N-H (2.68)	br d	d	m	dd	dd

3α,4α-Dihydroxy-5β-(aminomethyl)cyclohex-1-ene-1-carboxylic acid **(181)**

The ester carbamate (**179**) (61 mg, 0.20 mmol) was treated with trifluoroacetic acid (4 cm^3), the solution stirred at 20°C for 5 min and then concentrated under reduced pressure. The residue was dissolved in water (4 cm^3) and a 1.0 M aqueous sodium hydroxide solution was added dropwise until the solution was alkaline and then a further 0.3 cm^3 was added. The solution was stirred at 20°C under an argon atmosphere for 20 h. Amberlite IR 120 (+) ion exchange resin was added and the mixture filtered. The resin was washed with water (3 x 10 cm^3), the washings discarded and the product washed from the resin with dilute aqueous ammonia

solution (3 x 8 cm^3). Lyophilisation yielded the title compound as an off-white powder (30.5 mg, 80%): R_F 0.55 (reverse phase silica, water); $\delta_H(D_2O)$ 2.02 (1H, dd m, J_{gem} 17.0, $J_{6\beta,5}$ 11.0 Hz, 6β-H), 2.15 (1H, m, 5-H), 2.59 (1H, dd, J_{gem} 17.0, $J_{6\alpha,5}$ 4.5 Hz, 6α-H), 3.03 (1H, dd, J_{gem} 13.0, $J_{1',5}$ 6.0 Hz, 1′-H), 3.24 (1H, dd, J_{gem} 13.0, $J_{1',5}$ 7.0 Hz, 1′-H), 3.72 (1H, dd, $J_{4,5}$ 10.8, $J_{4,3}$ 4.0 Hz, 4-H), 4.25 (1H, dd, $J_{3,4}$ 4.0, $J_{3,2}$ 5.0 Hz, 3-H), 6.49 (1H, m, 2-H); $\delta_C(D_2O)$ 28.7 (C-6), 32.2 (C-5), 42.0 (C-1′), 65.2 (C-4), 71.8 (C-3), 129.6 (C-2), 137.3 (C-1), 175.0 (C=O); m/z (C.I., NH_3) 188 (MH^+, 100%), 172 (35), 170 (23), 152 (44), 126 (25), 108 (14).

3β,4β-Dihydroxy-5β-(aminomethyl)cyclohex-1-ene-1-carboxylic acid (182)

The ester carbamate (**180**) (93 mg, 0.31 mmol) was treated with trifluoroacetic acid (5 cm^3), the solution stirred at 20°C for 10 min and then concentrated under reduced pressure. The residue was dissolved in water (5 cm^3) and a 1.0 M aqueous sodium hydroxide solution was added dropwise until the solution was alkaline and then a further 0.4 cm^3 was added. The solution was stirred at 20°C under an argon atmosphere for 20 h. Amberlite IR 120 (+) ion exchange resin was added and the mixture filtered. The resin was washed with water (3 x 10 cm^3), the washings discarded and the product washed from the resin with dilute aqueous ammonia solution (3 x 9 cm^3). Lyophilisation yielded the title compound as an off-white powder (34 mg, 58%): R_F 0.55 (reverse phase silica, water); $\delta_H(D_2O)$ 2.14 (2H, m, 5-H, 6β-H), 2.41 (1H, d m, J_{gem} 16.0 Hz, 6α-H), 3.13 (1H, dd, J_{gem} 13.0, $J_{1',5}$ 6.8 Hz, 1′-H), 3.24 (1H, dd, J_{gem} 13.0, $J_{1',5}$ 6.0 Hz, 1′-H), 4.04 (1H, m, 4-H), 4.44 (1H, m, 3-H), 6.35 (1H, br s, 2-H); $\delta_C(D_2O)$ 24.1 (C-6), 35.0 (C-5), 41.3 (C-1′), 67.3 (C-4), 68.2 (C-3), 132.3 (C-2), 135.1 (C-1), 175.0 (C=O); m/z (C.I., NH_3) 188 (MH^+, 100%), 170 (62), 154 (40), 126 (68), 108 (28).

Methyl 3α,4α-dihydroxy-5β-[N-(trifluoroacetyl)aminomethyl]cyclohex-1-ene-1-carboxylate (189)

The carbamate (**179**) (3.85 g, 12.8 mmol) was added slowly to trifluoroacetic acid (45 cm^3). After stirring at 20°C for 30 min the reaction mixture was concentrated under reduced pressure and taken up in pyridine (50 cm^3). The solution was treated with trifluoroacetic anhydride (9.60 cm^3, 68.0 mmol), DMAP (catalytic) and stirred at 20°C for 3 days. The reaction mixture was then concentrated under reduced pressure and poured into water (100 cm^3). Dilute aqueous hydrochloric acid (200 cm^3) was added and the mixture extracted with ethyl acetate (3 x 150 cm^3). The combined extracts were dried (Na_2SO_4) and concentrated under reduced pressure. Column chromatography (petrol-ethyl acetate 1:3) yielded the title compound as a colourless crystalline solid (2.90 g, 76%): m.p. 116-117°C (from petrol-ethyl acetate); R_F 0.33 (petrol-ethyl acetate 1:3); (Found: C, 44.5; H, 4.8; N, 4.65. $C_{11}H_{14}F_3NO_5$ requires C, 44.4; H, 4.75; N, 4.7%); ν_{max}(nujol) 3400 (OH), 3310 (OH), 1735 (C=O), 1705 (C=O), 1655 (C=C), 1260, 1235, 1205, 1175 cm^{-1}; δ_H(D_6-Acetone) 2.00 (1H, dddd, J_{gem} 18.0, $J_{6\beta,5}$ 9.5, $J_{6\beta,2}$ 2.4, $J_{6\beta,3}$ 1.2 Hz, 6β-H), 2.20 (1H, m, 5-H), 2.58 (1H, br dd, J_{gem} 18.0, $J_{6\alpha,5}$ 5.0 Hz, 6α-H), 3.45 (1H, m, 1′-H), 3.60 (2H, m, 4-H, 1′-H), 3.71 (3H, s, OMe), 3.90 (2H, br s, 2 x OH), 4.26 (1H, br t, $J_{3,4}$ 4.5, $J_{3,2}$ 4.5 Hz, 3-H), 6.82 (1H, ddd, $J_{2,3}$ 4.5, $J_{2,6\beta}$ 2.4, $J_{2,6\alpha}$ 1.0 Hz, 2-H), 8.50 (1H, br s, NH); δ_C(D_6-Acetone) 27.7 (C-6), 35.3 (C-5), 41.8 (C-1′), 51.4 (OMe), 65.5 (C-4), 71.1 (C-3), 116.5 (q, $J_{C,F}$ 268 Hz, CF_3), 131.3 (C-1), 137.2 (C-2), 157.4 (C=O), 166.9 (C=O); m/z (C.I., NH_3) 315 (MNH_4^+, 100%), 297 (M^+, 38), 280 (16), 264 (17), 248 (11).

Homonuclear decoupling experiment data for (**189**):

Signal Irradiated (Chemical shift, δ)	Observed Resonance			
	2-H	3-H	6α-H	6β-H
Original resonance	ddd	br dd	br dd	dddd
3-H (4.26)	dd		ddd 18.0, 5.0, 1.0 Hz	ddd
6α-H (2.58)	dd	ddd 4.5, 4.5, 1.2 Hz		ddd

Methyl 3α,4α-dihydroxy-5β-(acetoxymethyl)cyclohex-1-ene-1-carboxylate (**190**)

A solution of the trifluoroacetamide (**189**) (0.38 g, 1.28 mmol) in acetic anhydride (100 cm^3) and glacial acetic acid (50 cm^3), was cooled to -4°C under a nitrogen atmosphere. Sodium nitrite (0.88 g, 12.8 mmol) was added, and the reaction mixture was stirred at -4 to 10°C for 17 h. The solution was concentrated under reduced pressure and poured into water (50 cm^3). Saturated brine (50 cm^3) was added and the mixture extracted with ethyl acetate (3 x 100 cm^3). The combined extracts were dried (Na_2SO_4) and concentrated under reduced pressure (azeotroping with toluene). Column chromatography (petrol-ethyl acetate 1:3) yielded the title compound as a yellow oil (98 mg, 31%): R_F 0.43 (petrol-ethyl acetate 1:3); $\nu_{max}(CHCl_3)$ 3450 (OH), 1710 (C=O), 1655 (C=C), 1365, 1230, 1070, 955, 910 cm^{-1}; $\delta_H(CDCl_3)$ 2.10 (3H, s, Me), 2.18 (2H, m, 5-H, 6β-H), 2.64 (1H, m, 6α-H), 2.73 (1H, br s, OH), 3.21 (1H, br s, OH), 3.58 (1H, dd, $J_{4,5}$ 9.5, $J_{4,3}$ 4.5 Hz, 4-H), 3.77 (3H, s, OMe), 4.15 (1H, dd, J_{gem} 11.4, $J_{1',5}$ 3.4 Hz, 1'-H), 4.34 (1H, dd, $J_{3,2}$ 5.0, $J_{3,4}$ 4.5 Hz, 3-H), 4.43 (1H, dd, J_{gem} 11.4, $J_{1',5}$ 4.4 Hz, 1'-H), 6.90 (1H, ddd, $J_{2,3}$ 5.0, $J_{2,6\beta}$ 2.0, $J_{2,6\alpha}$ 1.0 Hz, 2-H); $\delta_C(CDCl_3)$ 20.8 (Me), 27.0 (C-6), 34.6 (C-5), 52.0 (OMe), 64.7 (C-1'), 65.3 (C-4), 69.2 (C-3), 132.2 (C-1), 135.8 (C-2), 167.0 (C=O), 171.8 (C=O); m/z (C.I.) 245

(MH^+, 1%), 227 (10), 211 (5), 195 (12), 167 (100), 151 (20), 137 (15).

***Methyl 3β,4β-diacetoxy-5β-[N-(t-butoxycarbonyl)aminomethyl]cyclohex-1-ene-1-carboxylate* (192)**

A solution of the diol (**180**) (9.90 g, 32.9 mmol) in dichloromethane (100 cm^3) was treated with triethylamine (11.5 cm^3, 82.3 mmol), acetic anhydride (7.76 cm^3, 82.3 mmol) and DMAP (catalytic). The reaction mixture was stirred at 20°C under a nitrogen atmosphere for 21 h. The solution was diluted with dichloromethane (100 cm^3), washed with dilute aqueous hydrochloric acid (2 x 100 cm^3), saturated aqueous sodium bicarbonate solution (100 cm^3), dried ($MgSO_4$) and concentrated under reduced pressure. Column chromatography (petrol-ethyl acetate 7:3) yielded the title compound as a colourless oil which crystalised on standing (9.00 g, 71%): m.p. 94-96°C (from petrol-ethyl acetate); R_F 0.70 (petrol-ethyl acetate 1:1); (Found : C, 56.1; H, 7.1; N, 3.6. $C_{18}H_{27}NO_8$ requires C, 56.1; H, 7.1; N, 3.6%); $\nu_{max}(CHCl_3)$ 3420 (NH), 1710 br (C=O), 1425, 1365, 1155, 1085, 1015, 930 cm^{-1}; $\delta_H(CDCl_3)$ 1.44 (9H, s, C*Me*$_3$), 2.06 (3H, s, Me), 2.08 (2H, m, 5-H, 6β-H), 2.09 (3H, s, Me), 2.47 (1H, d m, J_{gem} 17.0 Hz, 6α-H), 2.85 (1H, m, 1′-H), 3.28 (1H, m, 1′-H), 3.78 (3H, s, OMe), 4.95 (1H, br t, *J* 6.0 Hz, NH), 5.45 (1H, m, 4-H), 5.57 (1H, m, 3-H), 6.64 (1H, m, 2-H); $\delta_C(CDCl_3)$ 20.7 (2 x Me), 24.4 (C-6), 28.3 (C*Me*$_3$), 36.4 (C-5), 41.2 (C-1′), 52.0 (OMe), 66.3 (C-4), 69.3 (C-3), 79.5 (*C*Me$_3$), 132.0 (C-1), 134.4 (C-2), 155.8 (C=O), 166.2 (C=O), 170.0 (C=O), 171.1 (C=O); m/z (C.I.) 386 (MH^+, 1%), 368 (1), 330 (55), 286 (50), 270 (20), 252 (10), 226 (100).

***Methyl 3β,4β-diacetoxy-5β-[N-(trifluoroacetyl)aminomethyl]cyclohex-1-ene-1-carboxylate* (193)**

The carbamate (**192**) (1.05 g, 2.72 mmol) was added portionwise to trifluoroacetic

acid (10 cm^3). After stirring for 10 min at 20°C the reaction mixture was concentrated under reduced pressure, taken up in pyridine (8 cm^3). Trifluoroacetic anhydride (0.37 cm^3, 3.90 mmol) and DMAP (catalytic) were added and the reaction mixture stirred at 20°C for 1.5 h. After diluting with ethyl acetate (200 cm^3), the solution was washed with dilute aqueous hydrochloric acid (2 x 100 cm^3), saturated aqueous sodium bicarbonate solution (100 cm^3), dried ($MgSO_4$) and concentrated under reduced pressure. Column chromatography (petrol-ethyl acetate 7:3) yielded the title compound as a colourless solid (1.01 g, 97%): m.p. 117-118°C (from petrol-ethyl acetate); R_F 0.70 (petrol-ethyl acetate 1:1); (Found : C, 47.0; H, 4.6; N, 3.6. $C_{15}H_{18}F_3NO_7$ requires C, 47.25; H, 4.8; N, 3.7%); ν_{max}(nujol) 3280 (NH), 1710 (C=O), 1270, 1230, 1150, 1050, 1025 cm^{-1}; $\delta_H(CDCl_3)$ 2.08 (3H, s, Me), 2.12 (1H, m, 6β-H), 2.13 (3H, s, Me), 2.30 (1H, m, 5-H), 2.54 (1H, br dd, J_{gem} 17.5, $J_{6\alpha,5}$ 5.5 Hz, 6α-H), 3.00 (1H, m, 1′-H), 3.61 (1H, m, 1′-H), 3.79 (3H, s, OMe), 5.37 (1H, m, 4-H), 5.58 (1H, m, 3-H), 6.67 (1H, br s, 2-H), 7.32 (1H, br t, J 6.0 Hz, NH); m/z (C.I.) 382 (MH^+, 3%), 322 (100), 262 (15), 230 (29).

***Methyl 3β,4β-diacetoxy-5β-(acetoxymethyl)cyclohex-1-ene-1-carboxylate* (194)**

A solution of the trifluoroacetamide (**193**) (5.00 g, 13.1 mmol) in glacial acetic acid (120 cm^3) and acetic anhydride (240 cm^3) was cooled to -4°C under a nitrogen atmosphere. Sodium nitrite (9.06 g, 131 mmol) was added and the solution was stirred at 0°C for 18 h. The reaction mixture was concentrated under reduced pressure and partitioned between ethyl acetate (400 cm^3) and water (150 cm^3). The aqueous phase was extracted with ethyl acetate (2 x 200 cm^3) and the combined organic extracts dried (Na_2SO_4), concentrated under reduced pressure and azeotroped with toluene. Column chromatography (petrol-ethyl acetate 4:1 to 7:3) yielded the title compound as a colourless oil (0.48 g, 11%): R_F 0.43 (petrol-ethyl acetate 7:3); ν_{max}(liquid film) 1740 (C=O), 1650 (C=C), 1430, 1370, 1240, 1150,

1090, 1040 cm^{-1}; $\delta_H(CDCl_3)$ 2.05 (3H, s, Me), 2.07 (3H, s, Me), 2.08 (3H, s, Me), 2.18 (1H, m, 6β-H), 2.33 (1H, m, 5-H), 2.50 (1H, br dd, J_{gem} 17.5, $J_{6\alpha,5}$ 5.5 Hz, 6α-H), 3.79 (3H, s, OMe), 3.97 (1H, dd, J_{gem} 11.2, $J_{1',5}$ 6.3 Hz, 1'-H), 4.03 (1H, dd, J_{gem} 11.2, $J_{1',5}$ 8.2 Hz, 1'-H), 5.51 (1H, m, 4-H), 5.60 (1H, m, 3-H), 6.65 (1H, br s, 2-H); $\delta_C(CDCl_3)$ 20.7 (2 x Me), 20.8 (Me), 23.4 (C-6), 52.0 (OMe), 63.4 (C-1'), 65.4 (C-4), 68.9 (C-3), 131.7 (C-1), 134.4 (C-2), 166.1 (C=O), 170.0 (C=O), 170.4 (C=O), 170.8 (C=O), m/z (C.I.) 329 (MH^+, 3%), 269 (100), 227 (6), 211(6), 209 (4), 195 (3).

Methyl 3β,4β-dihydroxy-5β-(hydroxymethyl)cyclohex-1-ene-1-carboxylate (**195**)

(a) A solution of the triacetate (**194**) (420 mg, 1.28 mmol) in methanol (25 cm^3) was treated with aqueous ammonia solution (8 cm^3) and stirred at 20°C for 2 days. The solvents were evaporated under reduced pressure. Column chromatography (ethyl acetate) yielded the title compound as a colourless powder (152 mg, 59%): m.p. 127-128°C (from ethyl acetate); R_F 0.28 (ethyl acetate); (Found : C, 53.6; H, 6.7. $C_9H_{14}O_5$ requires C, 53.5; H, 7.0%); ν_{max}(nujol) 3320 (OH), 3220 (OH), 1710 (C=O), 1640 (C=C), 1255, 1235, 1150, 1110, 1055, 1035, 970 cm^{-1}; $\delta_H(D_2O)$ 1.97 (2H, m, 5-H, 6β-H), 2.35 (1H, br d, 6α-H), 3.58 (1H, dd, J_{gem} 11.0, $J_{1',5}$ 6.0 Hz, 1'-H), 3.70 (1H, dd, J_{gem} 11.0, $J_{1',5}$ 7.2 Hz, 1'-H), 3.76 (3H, s, OMe), 4.08 (1H, m, 4-H), 4.42 (1H, m, 3-H), 6.68 (1H, br s, 2-H); $\delta_C(D_2O)$ 22.2 (C-6), 38.3 (C-5), 52.0 (OMe), 62.1 (C-1'), 66.2 (C-4), 68.7 (C-3), 130.0 (C-1), 138.8 (C-2), 168.8 (C=O); m/z (C.I.) 185 (M^+-OH, 70%), 167 (39), 153 (100), 137 (62).

(b) A solution of the trifluoroacetamide (**193**) (0.70 g, 1.84 mmol) in glacial acetic acid (70 cm^3) and acetic anhydride (140 cm^3) was cooled to -4°C under a nitrogen atmosphere. Sodium nitrite (1.27 g, 18.4 mmol) was added and the solution was stirred at -4 to 10°C for 18 h. The reaction mixture was concentrated under reduced pressure and poured into water (50 cm^3). Brine (50 cm^3) was added and the mixture

extracted with ethyl acetate (3 x 100 cm^3). The combined organic phases were dried (Na_2SO_4) and concentrated under reduced pressure (azeotroping with toluene) to leave a yellow oil (0.70 g). This crude product was dissolved in methanol (40 cm^3) and treated with dilute aqueous ammonia solution. After stirring at 20°C for 19 h the reaction mixture was concentrated under reduced pressure. Column chromatography (ethyl acetate) yielded the title compound as a pale yellow solid (0.18 g, 48%), identical with the sample described above.

***3β,4β-Dihydroxy-5β-(hydroxymethyl)cyclohex-1-ene-1-carboxylic acid* (196)**

A solution of the methyl ester (**195**) (81 mg, 0.40 mmol) in water (8 cm^3) was treated with aqueous sodium hydroxide solution (1.0 M, 0.42 cm^3) and stirred at 20°C under a nitrogen atmosphere for 18 h. The solution was acidified by adding Amberlite IR 120 (+) ion exchange resin and filtered. The resin was washed with water (3 x 5 cm^3) and the combined filtrate and washings lyophilised to yield the title compound as an off-white solid (75 mg, quantitative): m.p. 192-193°C (from acetonitrile); R_F 0.72 (reverse phase silica , water); $\delta_H(D_2O)$ 1.94 (2H, m, 5-H, 6β-H), 2.31 (1H, br d, J_{gem} 16.0 Hz, 6α-H), 3.57 (1H, dd, J_{gem} 11.0, $J_{1',5}$ 6.0 Hz, 1′-H), 3.69 (1H, dd, J_{gem} 11.0, $J_{1',5}$ 7.5 Hz, 1′-H), 4.06 (1H, m, 4-H), 4.41 (1H, m, 3-H), 6.65 (1H, br s, 2-H); $\delta_C(D_2O)$ 22.2 (C-6), 38.3 (C-5), 62.1 (C-1′), 66.2 (C-4), 68.7 (C-3), 130.6 (C-1), 138.5 (C-2), 170.4 (C=O); m/z (C.I.) 189 (MH^+, 5%), 171 (46), 153 (100), 137 (49).

***Methyl 3α,4α-isopropylidenedioxy-5β-(fluoromethyl)cyclohex-1-ene-1-carboxylate* (197)**

A polythene reaction vessel fitted with a septum cap was charged with dichloromethane (4 cm^3) and morpholino-DAST (0.14 cm^3, 1.14 mmol) under a

nitrogen atmosphere and cooled to -78°C. A solution of the alcohol (**172**) (275 mg, 1.14 mmol) in dichloromethane (2 cm^3) was added via cannular and the solution stirred for 30 min before warming to 20°C and stirring for a further 2 days. Silica gel was added and the dichloromethane evaporated under reduced pressure. The pre-absorbed product was added to the top of a silica gel column and eluted with petrol-ethyl acetate (9:1) to yield the title compound as a colourless oil (180 mg, 65%): R_F 0.50 (petrol-ethyl acetate 4:1); $\nu_{max}(CHCl_3)$ 1715 (C=O), 1645 (C=C), 1430, 1370, 1220, 1160, 1090, 1050 cm^{-1}; $\delta_H(CDCl_3)$ 1.41 (3H, s, Me), 1.44 (3H, s, Me), 1.99 (1H, d m, $J_{5,F}$ 25.6 Hz, 5-H), 2.15 (1H, ddt, J_{gem} 17.3, $J_{6\beta,5}$ 9.7, $J_{6\beta,2}$ 1.9, $J_{6\beta,3}$ 1.9 Hz, 6β-H), 2.70 (1H, br dd, J_{gem} 17.3, $J_{6\alpha,5}$ 4.1 Hz, 6α-H), 3.78 (3H, s, OMe), 4.16 (1H, dd, $J_{4,5}$ 9.0, $J_{4,3}$ 6.0 Hz, 4-H), 4.54 (1H, ddd, $J_{1',F}$ 49.0, J_{gem} 9.5, $J_{1',5}$ 4.0 Hz, 1′-H), 4.59 (1H, ddd, $J_{1',F}$ 49.0, J_{gem} 9.5, $J_{1',5}$ 5.1 Hz, 1′-H), 4.65 (1H, m, 3-H), 6.94 (1H, ddd, $J_{2,3}$ 3.5, $J_{2,6\beta}$ 1.9, $J_{2,6\alpha}$ 1.0 Hz, 2-H); $\delta_C(CDCl_3)$ 24.4 (d, $J_{6,F}$ 4 Hz, C-6), 25.7 (Me), 28.1 (Me), 38.3 (d, $J_{5,F}$ 18 Hz, C-5), 52.1 (OMe), 71.1 (C-4), 72.9 (d, $J_{3,F}$ 4 Hz, C-3), 83.7 (d, $J_{1',F}$ 170 Hz, C-1′), 109.2 (CMe_2), 133.1 (C-1), 133.7 (C-2), 166.6 (C=O); m/z (E.I.) 244 (M^+, 1%), 229 (M^+-CH_3, 229.0867 $C_{12}H_{17}FO_4$ requires 229.0876, 55%), 187 (16), 169 (28).

Further elution with petrol-ethyl acetate (4:6) yielded the unreacted alcohol (**172**) (65 mg, 23%).

***Methyl 3α,4α-isopropylidenedioxy-5β-formylcyclohex-1-ene-1-carboxylate* (198)**

A solution of the alcohol (**172**) (260 mg, 1.07 mmol) in dichloromethane (12 cm^3) was treated with pyridinium chlorochromate (0.35 g, 1.61 mmol) and stirred at 20°C for 4 h. The reaction mixture was added directly to the top of a silica gel column and eluted with petrol-ethyl acetate (7:3) to yield the title compound as a colourless oil (182 mg, 71%): R_F 0.55 (petrol-ethyl acetate 7:3); ν_{max}(liquid film) 1715 (C=O), 1650 (C=C), 1435, 1370, 1250, 1165, 1100, 1060, 1020, 860 cm^{-1}; $\delta_H(CDCl_3)$ 1.42

(3H, s, Me), 1.43 (3H, s, Me), 2.49 (1H, ddt, J_{gem} 18.0, $J_{6\beta,5}$ 7.0, $J_{6\beta,2}$ 1.6, $J_{6\beta,3}$ 1.6 Hz, 6β-H), 2.75 (1H, ddt, J_{gem} 18.0, $J_{6\alpha,5}$ 5.7, $J_{6\alpha,2}$ 1.5, $J_{6\alpha,3}$ 1.5 Hz, 6α-H), 2.93 (1H, ddd, $J_{5,6\beta}$ 7.0, $J_{5,4}$ 6.3, $J_{5,6\alpha}$ 5.7 Hz, 5-H), 3.78 (3H, s, OMe), 4.51 (1H, dd, $J_{4,5}$ 6.3, $J_{4,3}$ 6.3 Hz, 4-H), 4.72 (1H, m, 3-H), 6.89 (1H, ddd, $J_{2,3}$ 3.5, $J_{2,6\beta}$ 1.6, $J_{2,6\alpha}$ 1.5 Hz, 2-H), 9.79 (1H, d, J 0.5 Hz, CHO); $\delta_C(CDCl_3)$ 20.5 (C-6), 25.7 (Me), 27.7 (Me), 49.6 (C-5), 52.0 (OMe), 70.6 (C-4), 71.8 (C-3), 109.2 (CMe_2), 130.4 (C-1), 134.8 (C-2), 166.2 (C=O), 201.3 (CHO); m/z (E.I.) 241 (MH^+, 1%), 225 (M^+-CH_3, 225.0768 $C_{11}H_{13}O_5$ requires 225.0763, 21), 156 (100).

Homonuclear decoupling experiment data for (**198**):

Signal Irradiated (Chemical shift, δ)	**Observed Resonance**					
	2-H	3-H	4-H	5-H	6α-H	6β-H
Original resonance	ddd	m	dd	ddd	ddt	ddt
2-H (6.89)		d m 6.3 Hz	dd	ddd	ddd	ddd
3-H (4.72)	dd		m	ddd	ddd	ddd
4-H (4.51)	ddd	m simplifies		dd 5.7, 7.0 Hz	ddt	ddt
5-H (2.93)	ddd	m	d		dt	dt
6α-H (2.75)	dd	m simplifies	dd	m		m
6β-H (2.49)	dd	m simplifies	dd	m	m	

Methyl 3α,4α-isopropylidenedioxy-5β-(difluoromethyl)cyclohex-1-ene-1-carboxylate (**199**)

A polythene reaction vessel fitted with a septum cap was charged with dichloromethane (10 cm^3) and morpholino-DAST (0.11 cm^3, 0.91 mmol) under a

nitrogen atmosphere. A solution of the aldehyde (**198**) (182 mg, 0.76 mmol) in dichloromethane (4 cm^3) was added via cannular and the reaction mixture stirred at 20°C for 2 days. Silica gel was added and the dichloromethane evaporated under reduced pressure. The pre-absorbed product was added to the top of a silica gel column and eluted with petrol-ethyl acetate (9:1) to yield the title compound as a colourless oil (181 mg, 91%): R_F 0.74 (petrol-ethyl acetate 7:3); $\nu_{max}(CHCl_3)$ 1715 (C=O), 1645 (C=C), 1430, 1370, 1230, 1060, 910 cm^{-1}; $\delta_H(CDCl_3)$ 1.41 (3H, s, Me), 1.46 (3H, s, Me), 2.14 (2H, m, 5-H, 6β-H), 2.78 (1H, m, 6α-H), 3.79 (3H, s, OMe), 4.20 (1H, dd, $J_{4,3}$ 6.2, $J_{4,5}$ 9.0 Hz, 4-H), 4.66 (1H, m, 3-H), 6.02 (1H, td, $J_{1',F}$ 56.0 Hz, $J_{1',5}$ 2.0 Hz, 1'-H), 6.97 (1H, ddd, $J_{2,3}$ 3.5, $J_{2,6\beta}$ 2.0, $J_{2,6\alpha}$ 1.0 Hz, 2-H); $\delta_C(CDCl_3)$ 19.7 (t, $J_{6,F}$ 5 Hz, 6-H), 25.5 (Me), 27.9 (Me), 41.4 (t, $J_{5,F}$ 20 Hz, 5-H), 52.1 (OMe), 70.9 (C-4), 72.0 (d, $J_{3,F}$ 7 Hz, 3-H), 109.5 (CMe_2), 116.0 (t, $J_{1',F}$ 241 Hz, 1'-H), 132.4 (C-1), 133.3 (C-2), 166.2 (C=O); m/z (E.I.) 248 (M^+-CH_2, 248.0853 $C_{11}H_{14}F_2O_4$ requires 248.0860, 3%), 247 (M^+-CH_3, 39), 231 (3), 205 (5), 187 (20) (C.I.) 263 (MH^+, 100%), 247 (42), 205 (63), 187 (12).

Methyl 3α,4α-dihydroxy-5β-(fluoromethyl)cyclohex-1-ene-1-carboxylate **(200)**

A solution of the acetonide (**197**) (175 mg, 0.72 mmol) in THF (5 cm^3), glacial acetic acid (5 cm^3) and water (4 cm^3) was heated to 50-60°C under a nitrogen atmosphere for 2 days. The solution was concentrated under reduced pressure and chromatographed (petrol-ethyl acetate 1:3) to yield the title compound as a colourless oil which crystalised on standing (142 mg, 97%): m.p. 78-79°C (from petrol-ethyl acetate); R_F 0.25 (petrol-ethyl acetate 1:1); (Found : C, 52.95; H, 6.45. $C_9H_{13}FO_4$ requires C, 52.9; H, 6.4%); $\nu_{max}(CHCl_3)$ 3550 (OH), 3400 (OH), 1715 (C=O), 1650 (C=C), 1430, 1250, 1090, 1070, 965, 935, 905 cm^{-1}; $\delta_H(CDCl_3)$ 2.06-2.32 (2H, m, 6β-H), 2.47 (2H, br s, 2 x OH), 2.67 (1H, m, 6α-H), 3.77 (3H, s, OMe), 3.78 (1H, m, 4-H), 4.33 (1H, dd, $J_{3,4}$ 4.0, $J_{3,2}$ 5.0 Hz, 3-H), 4.55 (1H, ddd,

$J_{1',F}$ 47.0, J_{gem} 9.0, $J_{1',5}$ 3.8 Hz, 1′-H), 4.72 (1H, ddd, $J_{1',F}$ 47.0, J_{gem} 9.0, $J_{1',5}$ 4.0 Hz, 1′-H), 6.91 (1H, ddd, $J_{2,3}$ 5.0, $J_{2,6\beta}$ 2.0, $J_{2,6\alpha}$ 1.0 Hz, 2-H); $\delta_C(CDCl_3)$ 26.2 (d, $J_{6,F}$ 7 Hz, C-6), 36.0 (d, $J_{5,F}$ 18 Hz, C-5), 52.1 (OMe), 65.5 (C-4), 69.0 (d, $J_{3,F}$ 4 Hz, C-3), 84.1 (d, $J_{1',F}$ 168 Hz, C-1′), 132.8 (C-1), 135.4 (C-2), 166.9 (C=O); m/z (E.I.) 204 (M^+, 2%), 189 (1), 172 (12), 152 (24), 128 (21), 96 (100).

3α,4α-Dihydroxy-5β-(fluoromethyl)cyclohex-1-ene-1-carboxylic acid (**201**)

A solution of the methyl ester (**200**) (68 mg, 0.33 mmol) in THF (5 cm^3) was treated with aqueous sodium hydroxide solution (1.0 M, 0.35 cm^3). Water (*ca.* 4 cm^3) was added to give a homogeneous solution, which was stirred at 20°C under a nitrogen atmosphere for 3 h. Amberlite IR 120 (+) ion exchange resin was added and the mixture filtered. The resin was washed with water and the combined filtrate and washings were concentrated under reduced pressure and lyophilised to yield the title compound as an off-white solid (59 mg, 93%): R_F 0.50 (reverse phase silica, water); ν_{max}(nujol) 3300 (OH), 3140 (OH), 1685 (C=O), 1650 (C=C), 1270, 1240, 1070, 1050, 950 cm^{-1}; $\delta_H(D_2O)$ 2.17 (2H, m, 5-H, 6β-H), 2.54 (1H, m, 6α-H), 3.75 (1H, dd, $J_{4,5}$ 10.3, $J_{4,3}$ 4.0 Hz, 4-H), 4.27 (1H, dd, $J_{3,4}$ 4.0, $J_{3,2}$ 5.0 Hz, 3-H), 4.55 (1H, ddd, $J_{1',F}$ 47.0, J_{gem} 9.3, $J_{1',5}$ 3.2 Hz, 1′-H), 4.66 (1H, ddd, $J_{1',F}$ 47.0, J_{gem} 9.3, $J_{1',5}$ 4.2 Hz, 1′-H), 6.70 (1H, d m, $J_{2,3}$ 5.0 Hz, 2-H); $\delta_C(D_2O)$ 26.0 (d, $J_{6,F}$ 4 Hz, 6-H), 34.8 (d, $J_{5,F}$ 18 Hz, 5-H), 64.9 (C-4), 68.2 (d, $J_{3,F}$ 4 Hz, 3-H), 84.3 (d, $J_{1',F}$ 163 Hz, 1′-H), 133.7 (C-1), 134.0 (C-2), 171.8 (C=O); m/z (E.I.) 190 (M^+, 190.0643 $C_8H_{11}FO_4$ requires 190.0641, 1%), 172 (19), 152 (30), 114 (38), 96 (100).

Methyl 3α,4α-dihydroxy-5β-(difluoromethyl)cyclohex-1-ene-1-carboxylate (**202**)

A solution of the acetonide (**199**) (178 mg, 0.68 mmol) in THF (5 cm^3), glacial acetic acid (5 cm^3) and water (4 cm^3) was heated at 60°C under a nitrogen

atmosphere for 3 days. The solution was concentrated under reduced pressure. Column chromatography (petrol-ethyl acetate 4:6) yielded the title compound as a colourless oil which crystallised on standing (148 mg, 98%): m.p. 100-101°C (from petrol-ethyl acetate); R_F 0.32 (petrol-ethyl acetate 1:1); (Found: C, 48.4; H, 5.5. $C_9H_{12}F_2O_4$ requires C, 48.65; H, 5.4%); ν_{max}(nujol) 3260 (OH), 1700 (C=O), 1640 (C=C), 1255, 1145, 1125, 1100, 1090, 1025, 955, 945 cm^{-1}; δ_H(D_6-acetone, D_2O) 2.26 (1H, dddd, J_{gem} 17.6, $J_{6\beta,5}$ 10.5, $J_{6\beta,2}$ 2.5, $J_{6\beta,3}$ 1.0 Hz, 6β-H), 2.46 (1H, m, 5-H), 2.61 (1H, br dd, J_{gem} 17.6, $J_{6\alpha,5}$ 5.3 Hz, 6α-H), 3.72 (1H, dd, $J_{4,5}$ 10.4, $J_{4,3}$ 4.0 Hz, 4-H), 3.77 (3H, s, OMe), 4.30 (1H, m, 3-H), 6.26 (1H, td, $J_{1',F}$ 57.0, $J_{1',5}$ 2.2 Hz, 1′-H), 6.89 (1H, ddd, $J_{2,3}$ 5.2, $J_{2,6\beta}$ 2.5, $J_{2,6\alpha}$ 1.1 Hz, 2-H); δ_C($CDCl_3$ + D_6-acetone) 21.2 (t, $J_{6,F}$ 5 Hz, C-6), 38.6 (t, $J_{5,F}$ 20 Hz, C-5), 51.7 (OMe), 64.7 (C-4), 67.7 (d, $J_{3,F}$ 9 Hz, C-3), 116.3 (t, $J_{1',F}$ 240 Hz, 1′-H), 131.3 (C-1), 135.3 (C-2), 166.4 (C=O); m/z (E.I.) 222 (M^+, 3%), 190 (15), 170 (28), 128 (24), 96 (100).

3α,4α-Dihydroxy-5β-(difluoromethyl)cyclohex-1-ene-1-carboxylic acid (203)

A solution of the methyl ester (202) (26 mg, 0.12 mmol) in water (3 cm^3) was treated with aqueous sodium hydroxide solution (1.0 M, 0.12 cm^3,) and stirred at 20°C for 5 h. Amberlite IR-120 (+) ion exchange resin was added and the mixture filtered. The resin was washed with water (3 x 3 cm^3) and the combined filtrate and washings were lyophilised to yield the title compound as a white solid (21 mg, 86%): R_F 0.35 (reverse phase silica, water); ν_{max}(nujol) 3350 (OH), 3240 (OH), 1680 (C=O), 1630 (C=C), 1260, 1230, 1135, 1095, 1020, 990 cm^{-1}; δ_H(400 MHz, D_2O) 2.28 (1H, br dd, J_{gem} 17.5, $J_{6\beta,5}$ 10.1 Hz, 6β-H), 2.40 (1H, m, 5-H), 2.60 (1H, dd, J_{gem} 17.5, $J_{6\alpha,5}$ 5.2 Hz, 6α-H), 3.86 (1H, dd, $J_{4,5}$ 10.6, $J_{4,3}$ 4.1 Hz, 4-H), 4.29 (1H, m, 3-H), 6.14 (1H, td, $J_{1',F}$ 57.0, $J_{1',5}$ 2.3 Hz, 1′-H), 6.84 (1H, d m, $J_{2,3}$ 4.1 Hz, 2-H); δ_C(D_2O) 21.6 (t, $J_{6,F}$ 5 Hz, C-6), 37.8 (t, $J_{5,F}$ 20 Hz, C-5), 64.9 (C-4), 67.7 (d, $J_{3,F}$ 7 Hz, C-3), 117.0 (t, $J_{1',F}$ 239 Hz, 1′-H), 131.6 (C-1), 135.6 (C-2), 170.4 (C=O);

m/z (C.I., NH_3) 226 (MNH_4^+, 100%), 210 (88), 208 (M^+, 13), 182 (23); (E.I.) 190 (M^+-H_2O, 190.0432 $C_8H_8F_2O_3$ requires 190.0442, 18%).

Methyl 3α,4α-isopropylidenedioxy-5β-[2-methoxycarbonyl-1-nitroprop-3-yl]-cyclohex-1-ene-1-carboxylate **(208)**

A solution of diisopropylamine (0.39 cm^3, 2.75 mmol) in THF (4 cm^3) and DMPU (2 cm^3) was cooled to -35°C under a nitrogen atmosphere. A solution of *n*-butyl lithium in hexanes (1.6 M, 2.75 mmol) was added, the reaction mixture stirred for 10 mins, cooled to -78°C and stirred for a further 20 mins. Methyl 3-nitropropanoate (0.13 cm^3, 1.25 mmol) was added and the solution stirred for a further 1 h. A solution of the iodide (**165**) (200 mg, 0.57 mmol) in THF (1 cm^3) and DMPU (0.5 cm^3) was added dropwise via cannular. After stirring at -78°C for 5 h, the solution was allowed to warm to 0°C and stirred for a further 8 h. The reaction mixture was poured into saturated aqueous ammonium chloride solution (2 cm^3), diluted with water (10 cm^3) and extracted with ethyl acetate (3 x 30 cm^3). The combined extracts were dried ($MgSO_4$) and concentrated under reduced pressure. Column chromatography (petrol-ethyl acetate 17:3 then 7:3) yielded starting material (**165**) (120 mg, 60%) and the title compound as a yellow oil (40 mg, 20%): R_F 0.33 (petrol-ethyl acetate 7:3); $\nu_{max}(CHCl_3)$ 1735 (C=O), 1715 (C=O), 1560, 1440, 1375, 1240 and 1050 cm^{-1}; m/z (C.I.) 258 (MH^+, 31%), 342 (M^+-CH_3, 100), 300 (82), 268 (65), 250 (61).

Methyl 3α,4α-isopropylidenedioxy-5β-[2-methoxycarbonylprop-1-en-3-yl]-cyclohex-1-ene-1-carboxylate **(209)**

A solution of the nitro compound (**208**) (40 mg, 0.11 mmol) in THF (2 cm^3) was treated with 1,8-diazabicyclo[5.4.0]undec-7-ene (0.20 cm^3, 0.13 mmol). The

solution was stirred at 20°C for 4 h, filtered and concentrated under reduced pressure. Column chromatography (petrol-ethyl acetate 4:1) yielded the title compound as a colourless oil (30 mg, 86%): R_F 0.58 (petrol-ethyl acetate 7:3); $\nu_{max}(CHCl_3)$ 1710 (C=O), 1440, 1375, 1310, 1230, 1145, 1100, 1050 cm^{-1}; $\delta_H(CDCl_3)$ 1.39 (3H, s, Me), 1.43 (3H, s, Me), 1.91 (1H, ddt, J_{gem} 17.5, $J_{6\beta,5}$ 7.0, $J_{6\beta,2}$ 1.6, $J_{6\beta,3}$ 1.6 Hz, 6β-H), 2.11 (2H, m, 5-H, 3′-H), 2.53 (1H, dd m, J_{gem} 17.5, $J_{6\alpha,5}$ 4.0 Hz, 6α-H), 2.71 (1H, dd, J_{gem} 18.0, $J_{3',5}$ 9.0 Hz, 3′-H), 3.75 (3H, s, Me), 3.76 (3H, s, Me), 3.98 (1H, dd, $J_{4,5}$ 7.0, $J_{4,3}$ 5.5 Hz, 4-H), 4.59 (1H, m, 3-H), 5.56 (1H, br s, 1′-H), 6.24 (1H, d, $J_{1',3'}$ 1.3 Hz, 1′-H), 6.88 (1H, m, 2-H); $\delta_C(CDCl_3)$ 25.8 (C-6), 26.2 (Me), 28.1 (Me), 33.8 (C-3′), 35.6 (C-5), 51.9 (OMe), 52.0 (OMe), 71.1 (C-4), 77.2 (C-3), 109.1 (CMe_2), 127.0 (C-1′), 132.5 (C-1), 134.0 (C-2), 138.2 (C-2′), 167.0 (C=O), 167.3 (C=O); m/z (E.I.) 310 (M^+, 310.1429 $C_{16}H_{22}O_6$ requires 410.1414, 1%), 295 (35), 221 (52), 203 (100), 189 (22), 171 (22).

Homonuclear decoupling experiment data for (**209**):

Signal irradiated (Chemical shift, δ)	**Observed resonance** 2-H	3-H	4-H	6α-H	6β-H
Original resonance	m	m	dd	dd m	dddd
2-H (6.88)		d 5.5 Hz	dd	br dd	ddd
3-H (4.59)	dd		d 7.0 Hz	ddd	ddd

Methyl 3α,4α-hydroxy-5β-[2-methoxycarbonylprop-1-en-3-yl]cyclohex-1-ene-1-carboxylate (**210**)

A solution of the acetonide (**209**) (30 mg, 0.097 mmol) in THF (1 cm^3), glacial acetic acid (1 cm^3) and water (1 cm^3) was heated to 50-60°C under a nitrogen

atmosphere for 36 h. The reaction mixture was poured into saturated aqueous sodium bicarbonate solution (3 cm^3) and extracted with dichloromethane (10, 2 x 5 cm^3). The combined extracts were dried (Na_2SO_4) and concentrated under reduced pressure. Column chromatography (petrol-ethyl acetate 1:1 to 3:7) yielded the title compound as a colourless solid (12 mg, 47%): R_F 0.40 (petrol-ethyl acetate 1:3); $\nu_{max}(CHCl_3)$ 3520 (OH), 3400 (OH), 1710 (C=O), 1660 (C=C), 1630 (C=C), 1440, 1330, 1220, 1150, 1090, 960 cm^{-1}; $\delta_H(CDCl_3)$ 1.93 (1H, dddd, J_{gem} 18.0, $J_{6\beta,5}$ 9.0, $J_{6\beta,2}$ 2.1, $J_{6\beta,3}$ 1.3 Hz, 6β-H), 2.15 (1H, m, 5-H), 2.29 (1H, dd, J_{gem} 13.7, $J_{3',5}$ 8.1 Hz, 3′-H), 2.54 (1H, br dd, J_{gem} 18.0, $J_{6\alpha,5}$ 5.0 Hz, 6α-H), 2.72 (1H, ddd, J_{gem} 13.7, $J_{3',5}$ 4.0, $J_{3',1'}$ 0.9 Hz, 3′-H), 2.82 (1H, br s, OH), 3.39 (1H, br s, OH), 3.50 (1H, dd, $J_{4,5}$ 10.0, $J_{4,3}$ 4.2 Hz, 4-H), 3.74 (3H, s, Me), 3.77 (3H, s, Me), 4.32 (1H, br t, $J_{3,4}$ 4.2, $J_{3,2}$ 4.2 Hz, 3-H), 5.68 (1H, d, $J_{1',3'}$ 0.9 Hz, 1′-H), 6.29 (1H, d, J 1.3 Hz, 1′-H), 6.87 (1H, ddd, $J_{2,3}$ 4.2, $J_{2,6\beta}$ 2.1, $J_{2,6\alpha}$ 1.0 Hz, 2-H); $\delta_C(CDCl_3)$ 28.5 (C-6), 33.5 (C-3′), 34.5 (C-5), 52.0 (OMe), 52.2 (OMe), 65.8 (C-4), 71.6 (C-3), 128.2 (C-1′), 132.6 (C-1), 135.9 (C-2), 137.6 (C-2′), 167.1 (C=O), 168.2 (C=O); m/z (C.I., NH_3) 288 (MNH_4^+, 82%), 271 (MH^+, 25), 253 (100), 238 (25), 221 (82).

and the lactone (**211**) as a colourless solid (3.5 mg, 15%): R_F 0.49 (petrol-ethyl acetate 1:3); $\nu_{max}(CHCl_3)$ 3550 (OH), 3350 (OH), 1710 (C=O), 1655 (C=C), 1635 (C=C), 1335, 1275, 1160, 1110, 1050, 955 cm^{-1}; $\delta_H(CDCl_3)$ 2.01 (1H, dddd, J_{gem} 18.0, $J_{6\beta,5}$ 9.5, $J_{6\beta,2}$ 2.5, $J_{6\beta,3}$ 1.2 Hz, 6β-H), 2.26-2.48 (2H, m, 5-H, 3′-H), 2.62 (1H, d, $J_{OH,3}$ 3.3 Hz, 3-OH), 2.79-2.92 (2H, m, 3′-H, 6α-H), 3.78 (3H, s, OMe), 4.23 (1H, dd, $J_{4,5}$ 11.0, $J_{4,3}$ 3.8 Hz, 4-H), 4.50 (1H, m, 3-H), 5.66 (1H, m, 1′-H), 6.48 (1H, m, 1′-H), 6.95 (1H, dd, $J_{2,3}$ 5.5, $J_{2,6\beta}$ 2.5 Hz, 2-H); $\delta_C(CDCl_3)$ 27.8 (C-5), 30.7 (C-6), 34.2 (C-3′), 52.2 (OMe), 63.5 (C-4), 82.3 (C-3), 129.1 (C-1′), 132.9 (C-1), 133.0 (C-2′), 134.2 (C-2), 164.8 (C=O), 166.4 (C=O); m/z (C.I., NH_3) 256 (MNH_4^+, 100%), 239 (MH^+, 42), 21 (12), 207 (9).

3α,4α-Dihydroxy-5β-[2-carboxyprop-1-en-3-yl]cyclohex-1-ene-1-carboxylic acid **(212)**

A solution of the diester (**210**) (11 mg, 0.041 mmol) in water (1 cm^3) and THF (0.5 cm^3) was treated with aqueous sodium hydroxide solution (1.0 M, 0.09 cm^3) and stirred at 20°C under a nitrogen atmosphere for 4 h. The solution was acidified by adding Amberlite IRA-120 (+) ion exchange resin and filtered. The resin was washed with water and the combined filtrate and washings were lyophilised to yield the title compound as a cream solid (8.5 mg, 86%): R_F 0.47 (ethyl acetate - acetic acid 98:2); $\delta_H(D_2O)$ 1.93 (1H, dd m, J_{gem} 18.0, $J_{6\beta,5}$ 7.0 Hz, 6β-H), 2.14 (2H, m, 3′-H, 5-H), 2.42 (1H, dd m, J_{gem} 18.0, $J_{6\beta,5}$ 3.5 Hz, 6α-H), 2.68 (1H, dd, J_{gem} 19.0, $J_{3',5}$ 9.0 Hz, 3′-H), 3.66 (1H, dd, $J_{4,5}$ 8.2, $J_{4,3}$ 4.0 Hz, 4-H), 4.32 (1H, dd, $J_{3,4}$ 4.0, $J_{2,3}$ 4.0 Hz, 3-H), 5.74 (1H, s, 1′-H), 6.24 (1H, s, 1′-H), 6.80 (1H, m, 2-H); $\delta_C(D_2O)$ 26.9 (C-3′), 32.9 (C-6), 33.6 (C-5), 65.1 (C-4), 71.2 (C-3), 128.0 (C-1′), 131.5 (C-1), 136.5 (C-2), 137.7 (C-2′), 171.0 (C=O); m/z (-ve FAB) 241 ($M-H^-$, 5%), 223 (6), 138 (9), 122 (10).

Thermal Diels-Alder Reaction Using Chiral Auxiliary

A solution of the dihydropyridine (**132**) (5.38 g, 29.7 mmol) and the N-acryloyl sultam (**214**) (4.00 g, 14.9 mmol) in toluene (100 cm^3) was heated to reflux, under a nitrogen atmosphere, for 2.5 days. Toluene was evaporated under reduced pressure to leave a yellow oil. Column chromatography (petrol-ethyl acetate 4:1 then 7:3) yielded the mixed adducts (5.72 g, 85%) as a colourless solid. The ^1H NMR spectrum indicated two *endo* adducts as the major products, in 1:1 ratio, and a minor amount of the *endo* adducts present, with a *endo:exo* ratio of approximately 10:1. Repeated column chromatography and recrystallisations from petrol-ethyl acetate yielded the pure endo adducts:

Endo A (**215**) (0.96 g, 14%): m.p. 238-240°C (dec.) (from petrol-ethyl acetate); R_F 0.55 (petrol-ethyl acetate 7:3); (Found: C, 61.4; H, 7.75; N, 6.3. $C_{23}H_{34}N_2SO_5$ requires C, 61.3; H, 7.6; N, 6.2%); $\nu_{max}(CHCl_3)$ 1680 (C=O), 1390, 1360, 1330, 1160, 1125, 1110 cm^{-1}; $\delta_H(CDCl_3)$ 0.96 (3H, s, Me), 1.11 (3H, s, Me), 1.44 and 1.47 (9H, 2 x s, CMe_3), 0.90-1.60 (8H, m, 2 x 8-H, 2 x 3′-H, 2 x 5′-H, 2 x 6′-H), 2.26 (1H, m, 4′-H), 2.77 (1H, m, 4-H), 2.93 (1H, m, 3-H), 3.23 (1H, m, 3-H), 3.45 (2H, m, 2 x 10′-H), 3.60 (1H, m, 7-H), 3.84 (1H, dd, *J* 7.2, 5.2 Hz, 2′-H), 4.89 and 5.04 (1H, 2 x m, 1-H), 6.30-6.60 (2H, m, 5-H, 6-H); m/z (C.I.) 451 (MH^+, 3%), 435 (4), 395 (100), 377 (3), 351 (28).

Endo B (**216**) (1.04 g, 16%): m.p. 230-232°C (dec.) (from petrol-ethyl acetate); R_F 0.50 (petrol-ethyl acetate 7:3); (Found: C, 61.1; H, 7.7; N, 6.2. $C_{23}H_{34}N_2SO_5$ requires C, 61.3; H, 7.6; N, 6.2%); $\nu_{max}(CHCl_3)$ 1680 (C=O), 1400, 1365, 1340, 1160, 1130, 1120 cm^{-1}; $\delta_H(CDCl_3)$ 0.96 and 0.99 (3H, 2 x s, Me), 1.20 (3H, s, Me), 1.44 and 1.47 (9H, 2 x s, CMe_3), 1.25-2.07 (9H, m, 2 x 8-H, 2 x 3′-H, 4′-H, 2 x 5′-H, 2 x 6′-H), 2.88 (2H, m, 3-H, 4-H), 3.22 (1H, m, 3-H), 3.46 (2H, m, 2 x 10′-H), 3.66 (1H, m, 7-H), 3.83 (1H, m, 2′-H), 4.98 and 5.12 (1H, 2 x m, 1-H), 6.12-6.56 (2H, m, 5-H, 6-H); m/z (C.I.) 451 (MH^+, 7%), 435 (5), 395 (100), 377 (4), 351 (22).

***(-)-2-(t-Butoxycarbonyl)-7-endo-(methoxycarbonyl)-2-azabicyclo[2.2.2]oct-5-ene* (-)-(134)**

A solution of the endo adduct (**215**) (0.96 g, 2.13 mmol) in methanol (65 cm^3) was treated with potassium carbonate (0.96 g) and stirred at 20°C for 2.5 h. The reaction mixture was poured into water (200 cm^3), saturated brine added (100 cm^3) and extracted with dichloromethane (500, 2 x 200 cm^3). The combined extracts were dried ($MgSO_4$) and concentrated under reduced pressure. Column chromatography (petrol-ethyl acetate 4:1) yielded the title compound as a colourless solid (0.40 g, 70%): $[\alpha]_D^{20}$ -106° (*c* 1.02 in $CHCl_3$), spectral data identical to the racemic

compound (**134**) previously described. Continued elution (petrol-ethyl acetate 1:1) yielded the chiral auxiliary (**213**) as a colourless solid (0.35 g, 76%).

***(+)-2-(t-Butoxycarbonyl)-7-endo-(methoxycarbonyl)-2-azabicyclo[2.2.2]oct-5-ene* (+)-(134)**

The title compound was prepared from the endo adduct (**216**) using a similar procedure to that detailed above. The product was isolated as a colourless solid (0.45 g, 73%): $[\alpha]_D^{20}$ +104° (*c* 1.01 in $CHCl_3$), spectral data identical to the racemic compound (**134**) previously described. The chiral auxiliary (**213**) was recovered as a colourless solid (0.34g, 68%).

REFERENCES

REFERENCES

1. (a) E. Haslam, *The Shikimate Pathway*, Butterworths, London, 1974.
   (b) R. B. Herbert, *The Biosynthesis of Secondary Metabolites*, Chapman and Hall, London, 1981.
   (c) K. B. G. Torssell, *Natural Product Chemistry*, Wiley, Chichester, 1983.
   (d) B. Ganem, *Tetrahedron*, 1978, **34**, 3353.
2. (a) J. F. Eykmann, *Rec. Trav. Chim.*, 1885, **4**, 32.
   (b) J. F. Eykmann, *Ber. Dtsch. Chem. Ges.*, 1891, **24**, 1278.
3. (a) H. O. L. Fischer and G. Dangschat, *Helv. Chim. Acta.*, 1934, **17**, 1196.
   (b) H. O. L. Fischer and G. Dangschat, *Helv. Chim. Acta.*, 1935, **18**, 1204.
   (c) H. O. L. Fischer and G. Dangschat, *Helv. Chim. Acta.*, 1937, **20**, 705.
   (d) H. O. L. Fischer and G. Dangschat, *Ber. Dtsch. Chem. Ges.*, 1932, **65**, 1009.
4. K. Freudenberg, H. Meisenheimer, J. T. Lane and E. Plankenhorn, *Liebigs. Ann. Chem.*, 1940, ***543***, 162.
5. P. Karrer and K. P. Link, *Helv. Chim. Acta.*, 1927, **10**, 794.
6. B. D. Davis, *Adv. Enzymol.*, 1955, **16**, 287.
7. D. B. Sprinson, *Adv. Carbohydrate Chem.*, 1961, **15**, 235.
8. F. Gibson and J. Pittard, *Bact. Rev.*, 1968, **32**, 468.
9. J. R. Knowles, *Aldrichimica Acta*, 1989, **22**, 59.
10. P. A. Bartlett and K. Satake, *J. Am. Chem. Soc.*, 1988, **110**, 1628.
11. J. G. Levin and D. B. Sprinson, *J. Biol. Chem.*, 1964, **239**, 1142.
12. K. S. Anderson, J. A. Sikorski, A. J. Benesi and K. A. Johnson, *J. Am. Chem. Soc.*, 1988, **110**, 6577.
13. K. S. Anderson, R. D. Sammons, G. C. Leo, J. A. Sikorski, A. J. Benesi and K. A. Johnson, *Biochem.*, 1990, **29**, 1460.

14. P. N. Barlow, R. J. Appleyard, B. J. O. Wilson and J. N. S. Evans, *Biochem.*, 1989, **28**, 7985.

15. K. S. Anderson and K. A. Johnson, *Chem. Rev.*, 1990, **90**, 1131.

16. G. C. Leo, J. A. Sikorski and R. D. Sammons, *J. Am. Chem. Soc.*, 1990, **112**, 1653.

17. (a) D. K. Onderka and H. G. Floss, *J. Am. Chem. Soc.*, 1969, **91**, 5894.
(b) D. K. Onderka and H. G. Floss, *J. Biol. Chem.*, 1972, **247**, 736.

18. P. A. Bartlett, U. Maitra and P. M. Chouinard, *J. Am. Chem. Soc.*, 1986, **108**, 8068.

19. S. G. Sogo, T. S. Widlanski, J. H. Hoare, C. E. Grimshaw, G. A. Berchtold and J. R. Knowles, *J. Am. Chem. Soc.*, 1984, **106**, 2701.

20. R. McCridle, K. H. Overton and R. A. Raphael, *J. Chem. Soc.*, 1960, 1560.

21. S. A. Bowles, Ph.D. Thesis, University of Bath, 1988.

22. (a) M. M. Campbell, A. D. Kaye and M. Sainsbury, *Tetrahedron Lett.*, 1983, **24**, 4745.
(b) M. M. Campbell, A. D. Kaye, M. Sainsbury R. Yavarzadeh, *Tetrahedron Lett.*, 1984, **25**, 1629.
(c) M. M. Campbell, A. D. Kaye, M. Sainsbury R. Yavarzadeh, *Tetrahedron* 1984, **40**, 2461.

23. F. Brion, *Tetrahedron Lett.*, 1982, **23**, 5299.

24. O. Mitsunobu, *Synthesis*, 1981, 1.

25. M. M. Campbell, M. Sainsbury R. Yavarzadeh, *Tetrahedron* 1984, **40**, 5063.

26. D. Rajapaksa, B. A. Keay and R. Rodrigo, *Can. J. Chem.*, 1984, **62**, 826.

27. A. J. Birch, L. F. Kelly and D. V. Weerasuria, *J. Org. Chem.*, 1988, **53**, 278

28. B. M. R. Bandara, A. J. Birch and L. F. Kelly, *J. Org. Chem.*, 1984, **49**, 2496.

29. A. J. Birch and D. H. Williamson, *J. Chem. Soc., Perkin Trans. I*, 1973, 1892.

30. T. Takahashi, T. Namika, Y. Takeuchi and T. Koizumi, *Chem. Pharm. Bull.*, 1988, **36**, 3213.

31. H. Takayama, A. Iyobe and T. Koizumi, *J. Chem. Soc., Chem. Commun.*, 1986, 771.

32. P. X. Iten, A. A. Hofmann and C. H. Eugster, *Helv. Chim. Acta.*, 1978, **61**, 430.

33. M. Koreeda, K. Y. Jung and J. Ichita, *J. Chem. Soc., Perkin Trans. I*, 1989, 2129.

34. M. Koreeda, K. Teng and T. Murata, *Tetrahedron Lett.*, 1990, **31**, 5997.

35. M. Koreeda and M. A. Ciufolini, *J. Am. Chem. Soc.*, 1982, **104**, 2308.

36. I. Flemming and P. E. J. Sanderson, *Tetrahedron Lett.*, 1987, **28**, 4229.

37. (a) H. J. Bestmann and H. A. Heid, *Angew. Chem., Int. Ed. Engl.*, 1971, **10**, 336.

(b) M. Yoshikawa, Y. Ikeda, H. Kayakiri and I. Kitagawa, *Heterocycles*, 1982, **17**, 209.

(c) A. S. Mirza and A. Vasella, *Helv. Chim. Acta.*, 1984, **67**, 1562.

(d) T. Suami, K. Tadano, Y. Ueno and Y. Iimura, *Chem. Lett.*, 1985, 37.

38. (a) R. Grewe and E. Vangermain, *Chem. Ber.*, 1965, **98**, 104.

(b) G. Dangschat and H. O. L. Fischer, *Biochim. Biophys. Acta.*, 1950, **4**, 199.

(c) R. Grewe, H. Büttner and G. Burmeister, *Angew. Chem.*, 1957, **69**, 61.

(d) J. Cleofax, D. Mercier and S. D. Gero, *Angew. Chem., Int. Ed. Engl.*, 1971, **10**, 652.

(e) J. Cleofax, J. Leboul, D. Mercier, A. Gaudemer and S. D. Gero, *Bull. Soc. Chim. Fr.*, 1973, 2992.

39. G. W. J. Fleet, T. K. M. Shing and S. M. Warr, *J. Chem. Soc., Perkin Trans. I*, 1984, 905.

40. J. S. Brimacombe, F. Hunedy and L. C. N. Tucker, *J. Chem. Soc. C*, 1968, 1381.

41. S. Mirza and J. Harvey, *Tetrahedron Lett.*, 1991, **32**, 4111.

42. P. A. Bartlett and L. A. McQuaid, *J. Am. Chem. Soc.*, 1984, **106**, 7854.

43. K. E. Coblens, V. B. Muralidharan and B. Ganem, *J. Org. Chem.*, 1982, **47**, 5041.

44. E. Uhlmann and W. Pfleiderer, *Tetrahedron Lett.*, 1980, **21**, 1181.

45. D. A. McGowan and G. A. Berchtold, *J. Am. Chem. Soc.*, 1982, **104**, 7036.

46. B. D. Davis and E. S. Mingioli, *J. Bacteriol.*, 1953, **66**, 129.

47. C. P. Teng, Y. Yukimoto and B. Ganem, *Tetrahedron Lett.*, 1985, **26**, 21.

48. P. M. Chouinard and P. A. Bartlett, *J. Org. Chem.*, 1986, **51**, 75.

49. D. A. McGowan and G. A. Berchtold, *J. Am. Chem. Soc.*, 1982, **104**, 1153.

50. J. H. Hoare, P. P. Policastro and G. A. Berchtold *J. Am. Chem. Soc.*, 1983, **105**, 6264.

51. B. Ganem, N. Ikota, V. B. Muralidharan, W. S. Wade, S. D. Young and Y. Yukimoto, *J. Am. Chem. Soc.*, 1982, **104**, 6787.

52. (a) B. Ganem and N. Ikota, *J. Am. Chem. Soc.*, 1978, **100**, 351.
(b) B. Ganem and N. Ikota, *J. Chem. Soc., Chem Commun.*, 1978, 869.

53. D. Lesuisse and G. A. Berchtold, *J. Org. Chem.*, 1985, **50**, 888.

54. J. L. Pawlak and G. A. Berchtold, *J. Org. Chem.*, 1987, **52**, 1765.

55. H. B. Wood and B. Ganem, *J. Am. Chem. Soc.*, 1990, **112**, 8907.

56. S. Greenberg and J. G. Moffatt, *J. Am. Chem. Soc.*, 1973, **95**, 4016.

57. T. Fex, J. Trofast and B. Wickberg, *Acta. Chem. Scand.*, 1981, **35B**, 91.

58. H. B. Wood and B. Ganem, *Tetrahedron Lett.*, 1989, **30**, 6257.

59. T. D. Nelson and G. H. Posner, *Tetrahedron*, 1990, **46**, 4573.
See also: G. H. Posner, A. Haces, W. Harrison and C. M. Kinter, *J. Org. Chem.*, 1987, **52**, 4836.

60. J. H. Ottaway and D. K. Apps, *Biochemistry,* Bailliére Tindall, London, 1984, 4th Edn., p. 29-32.

61. H. C. Steinrücken and N. Amrheim, *Biochem. Biophys. Res. Commun.*,1980, **94**, 1207.

62. (a) D. G. Albert and P. A. Bartlett, *J. Am. Chem. Soc.*,1989, **111**, 2337.
(b) D. G. Albert, P. A. Bartlett, A. Fässler, C. B. Grisson, C. T. Lauhon, R. Nyfeler and K. L. McLaren, BCPC Monograph, 1989, **42**, 155

63. (a) R. Wolfenden, *Annu. Rev. Biophys. Bioeng.*, 1976, **5**, 271.
(b) P. A. Bartlett and G. R. Stark, *Pharmacol. Ther.*, 1984, **23**, 45.

64. R. Azerad, C. Froussios, M. Level and P. le Maréchal, *Biochem. Biophys. Res. Commun.*, 1980, **92**, 1104.

65. P. D. Pansegrau, K. S. Anderson, T. Widlanski, J. E. Ream, R. D. Sammons, J. A. Sikorski and J. R. Knowles, *Tetrahedron Lett.*, 1991, **32**, 2589.

66. D. A. McGowan and G. A. Berchtold, *J. Org. Chem.*, 1981, **46**, 2381.

67. For an attempted synthesis of this compound see:
G. M. Mandair, B.Sc. Final Year Project Report, University of Bath, 1991.

68. D. M. Stout and A. I. Meyers, *Chem. Rev.*, 1982, **82**, 223.

69. F. W. Fowler, *J. Org. Chem.*, 1972, **37**, 1321.

70. H. Sliwa and Y. Le Bot, *Tetrahedron Lett.*, 1977, **18**, 4129.

71. I. Fleming, *Frontier Orbitals and Organic Chemical Reactions,* Wiley, London, 1976.

72. K. Kobayashi, S. Miyazawa, A. Terahara, H. Mishima and H. Kurihura, *Tetrahedron Lett.*, 1976, **17**, 537.

73. R. J. Sundberg and J. D. Bloom, *J. Org. Chem.*, 1981, **46**, 4836.

74. T. W. Greene, *Protective Groups in Organic Synthesis*, Wiley, Chichester, 1981.

75. (a) R. J. Baumgarten, P. J. DeChristopher, J. P. Adamek, G. D. Lyon and S. A. Klein, *J. Org. Chem.*, 1974, **39**, 3525.

(b) P. J. DeChristopher, J. P. Adamek, G. D. Lyon, J. J. Galante, H. E. Haffner, R. J. Boggio and R. J. Baumgarten, *J. Am. Chem. Soc.*, 1969, **91**, 2384.

(c) Reviewed in: S. Patai, *The Chemistry of Functional Groups, Supplement F: The Chemistry of Amino, Nitroso and Nitro Compounds and their Derivatives*, Wiley, Chichester, 1982, Part 2, Ch. 22.

76. M. Schröder, *Chem Rev.*, 1980, **80**, 187.

77. V. VanRheenen, R. C. Kelly and D. Y. Cha, *Tetrahedron Lett.*, 1976, **17**, 1973.

78. M. Karplus, *J. Chem. Phys.*, 1959, **30**, 11.

79. For full discussions of the determination of solution conformations of shikimic acid by ^{1}H NMR spectroscopy see:

(a) L. D. Hall, *J. Org. Chem.*, 1964, **29**, 297.

(b) B. Talapartra, A. K. Das, S. K. Talapatra, *Phytochemistry*, 1989, **28**, 290.

(c) S. Castellino, G. C. Leo, R. D. Sammons and J. A. Sikorski, *J. Org. Chem.*, 1991, **56**, 5176.

80. (a) A. H. Haines, *Methods for the Oxidation of Organic Compounds - Alkanes, Alkenes, Alkynes and Arenes*, Academic Press, London, 1985, p. 86-88 and 96-98.

(b) H. O. House, *Modern Synthetic Reactions*, Benjamin, Menlo Park, 1972, 2nd Edn., p. 436-441.

81. R. B. Woodward and F. V. Brutcher, Jr., *J. Am. Chem. Soc.*, 1958, **80**, 209.

82. P. J. DeChristopher, J. P. Adamek, S. A. Klein, G. D. Lyon and R. J. Baumgarten, *J. Org. Chem.*, 1975, **40**, 3288.

83. M. Hiraoka, *Crown Compounds their characteristics and applications*, Elsevier, Oxford, 1982, p. 167-172 and references described therein.

84. N. H. Andersen, H. S. Uh, *Synthetic Commun.*, 1972, **2**, 297.

85. N. Nikolaides and B. Ganem, *J. Org. Chem.*, 1989, **54**, 5996.

86. N. Nikolaides and B. Ganem, *Tetrahedron Lett.*, 1989, **30**, 1461.

87. T. J. Lobl, *J. Chem. Educ.*, 1972, **49**, 730.

88. (a) W. J. Middleton, *J. Org. Chem.*, 1975, **40**, 574.
(b) *Aldrichchim Acta*, 1988, **21**, 27.

89. *Aldrichchim Acta*, 1988, **21**, 109.

90. G. Piancatelli, A. Scettri and M. D. Auria, *Synthesis*, 1982, 245.

91. S. V. Ley and W. P. Griffith, *Aldrichimica Acta*, 1990, **23**, 13.

92. For a listing of α-acrylate anion synthons, see Reference 94, and references therein.

93. D. Seebach, R. Henning and T. Mukhopadhyay, *Chem. Ber.*, 1982, **115**, 1705.

94. L. C. Yu and P. Helquist, *J. Org. Chem.*, 1981, **46**, 4536.

95. P. G. Baraldi, A. Barco, S. Benetti, F. Moroder, G. P. Pollini, D. Simoni and V. Zanirato, *J. Chem. Soc., Chem. Commun.*, 1982, 1265.

96. H. M. R. Hoffmann and J. Rabe, *Angew. Chem., Int. Ed. Engl.*, 1983, **22**, 795.

97. H. M. R. Hoffmann and J. Rabe, *Angew. Chem., Int. Ed. Engl.*, 1983, **22**, 796.

98. M. M. Campbell and H. S. Rabiasz, unpublished results.

99. (a) W. Caruthers, *Cycloaddition Reactions in Organic Synthesis*, Pergamon Press, Oxford, 1990.
(b) L. A. Paquette in *Asymmetric Synthesis*, Vol 3, Ch 4, Academic Press, 1984, Ed. J. D. Morrison, and references contained therein.

100. K. Narasaka, *Synthesis*, 1991, 1.

101. W. Oppolzer, *Tetrahedron*, 1987, **43**, 1969 and 4057

102. (a) M. R. Ashton, B.Sc. Final Year Project Report, University of Bath, 1990.
(b) M. A. Sage, B.Sc. Final Year Project Report, University of Bath, 1991.

103. D. A. Evans, K. T. Chapman and J. Bisaha, *J. Am. Chem. Soc.*, 1988, **110**, 1238.

104. T. Poll, A. Sobczak, H. Hartmann and G. Helmchen, *Tetrahedron Lett.*, 1985, **26**, 3095.

105. (a) W. Oppolzer, C. Chapuis and G. Bernardinelli, *Helv. Chim. Acta*, 1984, **67**, 1397.

106. B. H. Kim and J. Y. Lee, *Tetrahedron: Asymmetry*, 1991, **2**, 1359.

107. (a) M. Mehmandoust, C. Marazano, R. Singh, B. Gillet, M. Césario, J. L. Fourrey and B. C. Das, *Tetrahedron Lett.*, 1988, **29**, 4423.
(b) C. Marazano, S. Yannic, Y. Genisson, M. Mehmandoust and B. C. Das, *Tetrahedron Lett.*, 1990, **31**, 1995.

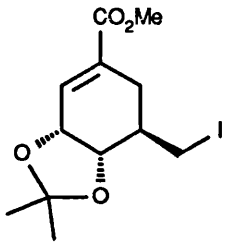
108. R. Daniels and J. L. Fischer, *J. Org. Chem.*, 1963, **28**, 320.

109. D. D. Perrin, W. L. F. Armarego and D. R. Perrin, *Purification of Laboratory Chemicals*, Pergamon, Oxford, 1980, 2nd Edn.

APPENDICES

APPENDIX ONE

X-ray Crystallographic Data for (165)



(165)

Compound (**165**) was crystallised from light petrol.

Crystal Data: $C_{12}H_{17}O_4I$, triclinic, a = 5.660(2), b = 8.252(4), c = 15.014(3)Å, $\underline{\alpha}$ = 97.32(3), $\underline{\beta}$ = 95.59(2), $\underline{\gamma}$ = 94.34(3)°, U = 689.4Å^3, space group P1, Z = 2, D_c = 1.687gcm^{-3}, F(000) = 344. Data were measured at room temperature on a Hilger and Watts Y290 four-circle diffractometer in the range $2<\theta<24°$. 2298 reflections were collected of which 1704 were unique with $I \geq 3\sigma(I)$. Data were corrected for Lorentz and polarization effects but not for absorption. The structure was solved by Patterson methods and refined using the SHELX[1] suite of programs. In the final least squares cycles the iodine and oxygen atoms were allowed to vibrate anisotropically. All other atoms were treated isotropically. Hydrogen atoms were included at calculated positions. Final residuals after 7 cycles of full-matrix least squares were $R = R_w$ = 0.0787 for unit weights. Maximum final shift/esd was 0.002, and the maximum and minimum residual densities were 0.84 and -0.80 eÅ^{-3} respectively. Final fractional atomic coordinates and isotopic thermal parameters, hydrogen atom positions, anisotropic thermal parameters, bond distances and bond angles are given below (**Tables 1 to 7**). The asymmetric unit is shown in **Figure 1**, along with the labelling scheme used.

1. (a) Sheldrick G.M., SHELX86, a computer program for crystal structure determination, University of Goetingen, 1986.

(b) Sheldrick G.M., SHELX76, a computer program for crystal structure determination, University of Cambridge, 1976.

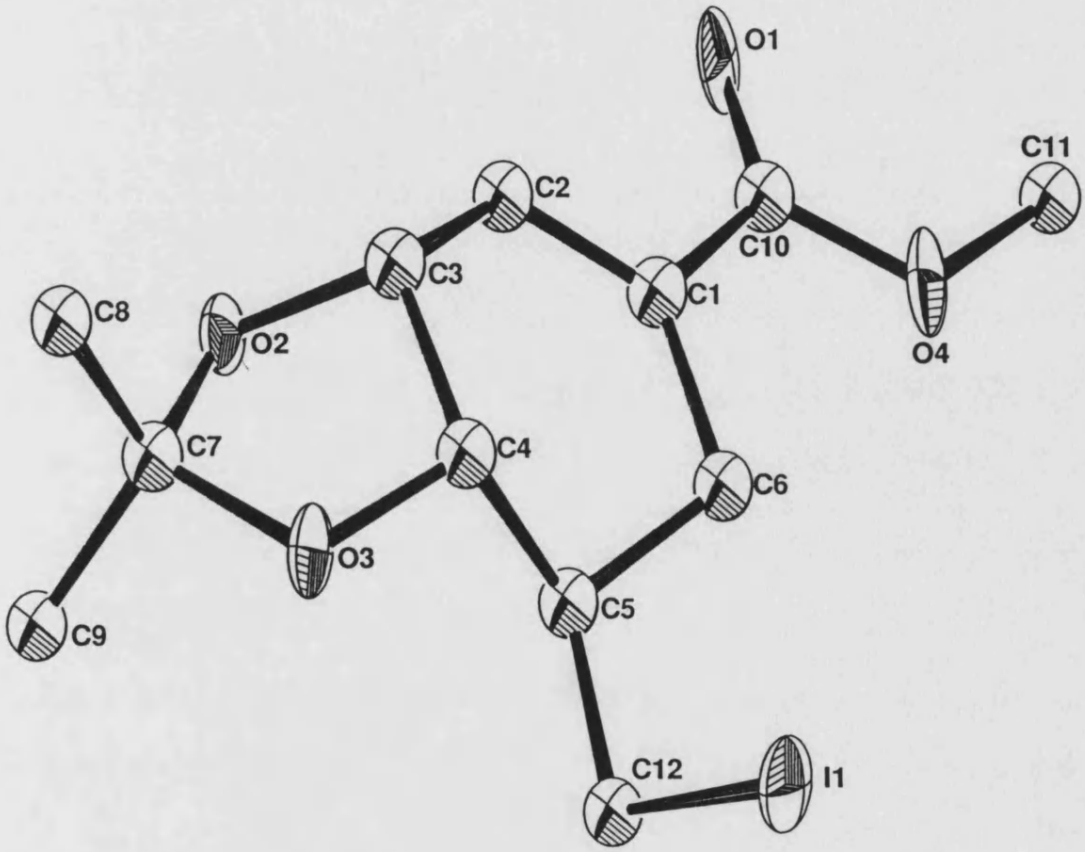


Figure 1

Table 1 Fractional atomic coordinates and thermal parameters (Å) for (165)

Atom	x	y	z	Uiso or Ueq (***)
I1	0.19475(2)	-0.74316(1)	0.02702(1)	0.0701(1) ***
O1	-0.36787(22)	-0.20378(12)	0.33775(6)	0.0732(7) ***
O2	0.15541(16)	-0.65099(10)	0.41271(5)	0.0461(5) ***
O3	0.30655(19)	-0.78459(11)	0.29193(5)	0.0605(6) ***
O4	-0.40734(21)	-0.26803(12)	0.18739(6)	0.0720(7) ***
C1	-0.1497(22)	-0.4120(14)	0.2705(8)	0.042(3)
C2	-0.0028(23)	-0.4221(15)	0.3483(9)	0.048(3)
C3	0.1912(22)	-0.5337(14)	0.3513(8)	0.041(3)
C4	0.2086(21)	-0.6429(14)	0.2631(8)	0.041(3)
C5	-0.0300(22)	-0.6837(14)	0.2060(8)	0.041(3)
C6	-0.1360(23)	-0.5249(15)	0.1839(8)	0.046(3)
C7	0.3021(22)	-0.7800(15)	0.3887(8)	0.043(3)
C8	0.5505(28)	-0.7440(19)	0.4354(10)	0.064(4)
C9	0.1880(30)	-0.9355(19)	0.4084(11)	0.069(4)
C10	-0.3115(23)	-0.2836(15)	0.2714(9)	0.046(3)
C11	-0.5802(32)	-0.1465(20)	0.1797(11)	0.074(4)
C12	-0.0337(25)	-0.8140(16)	0.1262(9)	0.051(3)

Table 2 Fractional atomic coordinates for the hydrogen atoms

Atom	x	y	z
H31	0.3475	-0.4486	0.3713
H41	0.3181	-0.5833	0.2196
H51	-0.1481	-0.7424	0.2470
H61	-0.3131	-0.5533	0.1489
H62	-0.0230	-0.4655	0.1414
H81	0.5475	-0.7411	0.5074
H82	0.6216	-0.6259	0.4212
H83	0.6615	-0.8372	0.4109
H91	0.2956	-1.0352	0.3912
H92	0.0171	-0.9582	0.3680
H93	0.1621	-0.9266	0.4791
H111	-0.6432	-0.1476	0.1093
H112	-0.4911	-0.0275	0.2065
H113	-0.7295	-0.1719	0.2169
H121	0.0260	-0.9236	0.1501
H122	-0.2139	-0.8393	0.0938

Table 3 Anisotropic thermal parameters (Å)

Atom	U11	U22	U33	U23	U13	U12
I1	0.096(1)	0.080(1)	0.034(1)	-0.005(1)	0.012(1)	0.028(1)
O1	0.100(8)	0.069(7)	0.051(6)	-0.005(5)	0.013(6)	0.044(6)
O2	0.062(5)	0.045(5)	0.031(4)	0.005(3)	0.010(4)	0.017(4)
O3	0.086(7)	0.064(6)	0.032(5)	0.004(4)	0.008(5)	0.045(6)
O4	0.093(8)	0.073(7)	0.050(6)	0.008(5)	0.005(6)	0.051(6)

Table 4 Bond lengths (Å)

I1	-C12	2.171(13)	O1	-C10	1.206(14)
O2	-C3	1.436(14)	O2	-C7	1.430(14)
O3	-C4	1.429(14)	O3	-C7	1.450(14)
O4	-C10	1.349(15)	O4	-C11	1.460(18)
C1	-C2	1.381(17)	C1	-C6	1.513(16)
C1	-C10	1.452(16)	C2	-C3	1.486(17)
C3	-C4	1.521(16)	C4	-C5	1.518(16)
C5	-C6	1.543(16)	C5	-C12	1.501(17)
C7	-C8	1.500(20)	C7	-C9	1.470(19)

Table 5 Bond angles (°)

C7	-O2	-C3	106.2(9)	C7	-O3	-C4	109.8(8)
C11	-O4	-C10	117(1)	C6	-C1	-C2	122(1)
C10	-C1	-C2	119(1)	C10	-C1	-C6	120(1)
C3	-C2	-C1	123(1)	C2	-C3	-O2	112(1)
C4	-C3	-O2	102.4(9)	C4	-C3	-C2	115(1)
C3	-C4	-O3	103.3(9)	C5	-C4	-O3	113(1)
C5	-C4	-C3	113(1)	C6	-C5	-C4	110(1)
C12	-C5	-C4	115(1)	C12	-C5	-C6	114(1)
C5	-C6	-C1	110(1)	O3	-C7	-O2	104.2(9)
C8	-C7	-O2	112(1)	C8	-C7	-O3	110(1)
C9	-C7	-O2	109(1)	C9	-C7	-O3	110(1)
C9	-C7	-C8	112(1)	O4	-C10	-O1	122(1)
C1	-C10	-O1	126(1)	C1	-C10	-O4	112(1)
C5	-C12	-I1	112.9(9)				

Table 6 Intermolecular distances (Å)

O1	...H31	2.62	1	1.0	0.0	0.0
O1	...H91	2.57	1	1.0	-1.0	0.0
O1	...H92	2.82	1	0.0	-1.0	0.0
O1	...H81	2.70	-1	0.0	-1.0	1.0
O1	...H93	2.92	-1	0.0	-1.0	1.0
O3	...H112	2.64	1	-1.0	1.0	0.0
C2	...H82	2.99	1	1.0	0.0	0.0
H31	...C10	2.92	1	-1.0	0.0	0.0
C9	...H93	2.98	-1	0.0	-2.0	1.0

Table 7 Intramolecular distances (Å)

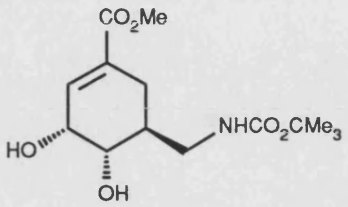
I1	...H121	2.72	I1	...H122	2.71
O1	...O4	2.24	O1	...C1	2.37
O1	...C2	2.85	O1	...C11	2.67
O1	...H112	2.66	O1	...H113	2.65
O2	...O3	2.27	O2	...C2	2.42
O2	...H31	2.12	O2	...C4	2.30
O2	...H51	2.86	O2	...C8	2.43
O2	...H81	2.72	O2	...H82	2.62
O2	...C9	2.36	O2	...H92	2.58
O2	...H93	2.60	O3	...C3	2.31
O3	...H31	2.86	O3	...H41	2.10
O3	...C5	2.45	O3	...H51	2.66
O3	...C8	2.41	O3	...H82	2.64
O3	...H83	2.65	O3	...C9	2.39

O3	...H91	2.70	O3	...H92	2.54
O3	...C12	2.97	O3	...H121	2.61
O4	...C1	2.32	O4	...C6	2.71
O4	...H61	2.46	O4	...H62	2.90
O4	...H111	2.08	O4	...H112	2.07
O4	...H113	2.11	C1	...C3	2.52
C1	...C4	2.89	C1	...C5	2.50
C1	...H51	2.70	C1	...H61	2.12
C1	...H62	2.14	C2	...H31	2.01
C2	...C4	2.53	C2	...C5	2.82
C2	...H51	2.89	C2	...C6	2.53
C2	...C10	2.44	C3	...H41	2.17
C3	...C5	2.53	C3	...H51	2.71
C3	...C6	2.99	C3	...C7	2.29
C3	...H82	2.76	H31	...C4	2.17
H31	...C7	2.77	H31	...C8	3.00
C4	...H51	2.10	C4	...C6	2.51
C4	...H62	2.77	C4	...C7	2.36
C4	...C12	2.55	C4	...H121	2.76
H41	...C5	2.06	H41	...C6	2.67
H41	...C12	2.78	C5	...H61	2.16
C5	...H62	2.15	C5	...H121	2.11
C5	...H122	2.11	H51	...C6	2.13
H51	...C12	2.02	C6	...C10	2.56
C6	...C12	2.56	C6	...H122	2.75
H61	...C10	2.70	H61	...C12	2.77
H62	...C10	2.99	H62	...C12	2.85
C7	...H81	2.12	C7	...H82	2.11

C7	...H83	2.13	C7	...H91	2.11
C7	...H92	2.07	C7	...H93	2.10
C8	...C9	2.46	C8	...H91	2.68
C8	...H93	2.75	H81	...C9	2.69
H83	...C9	2.74	C10	...C11	2.40
C10	...H112	2.66	C10	...H113	2.69

APPENDIX TWO

X-ray Crystallographic Data for (179)



(179)

Compound (**179**) was crystallised from ethyl acetate. A blocky crystal of approximate dimensions 0.25 x 0.25 x 0.25 mm was selected for data collection.

Crystal Data: $C_{14}H_{23}O_6N$, M = 302.34, monoclinic, a = 11.101(2), b = 14.136(3), c = 11.148(3) Å, β = 111.41(2), U = 1628.4 Å^3, space group $P2_1/a$, Z = 4, D_C = 1.12 gcm^{-3}, μ(Mo-K_α) = 0.60 cm^{-1}, F(000) = 648. Data were measured at room temperature on a Hilger and Watts Y290 four-circle diffractometer in the range $2<\theta<24°$. 2744 reflections were collected of which 1171 were unique with $I \geq 3\sigma(I)$. Data were corrected for Lorentz and polarization effects but not for absorption. The structure was solved by conventional direct methods.[1] In the latter stages of convergence the O3, O5, O6 and N1 atoms and carbons 9-13 were refined anisotropically. All other atoms were treated isotropically. The hydrogen atoms on the sp^2 hybridised carbon (C3) and on the nitrogen atom (N1) were located in the penultimate difference Fourier and these positions were refined with a constrained bond length of 1.08 Å in the final least squares cycles. The remaining hydrogens were included at calculated positions. Final residuals after 11 cycles of full-matrix least squares were $R = R_w$ = 0.0847 for unit weights. The total number of parameters varied was 141. Maximum final shift/esd was 0.004, and the maximum and

minimum residual densities were 0.17 and -0.12 $eÅ^{-3}$repectively. Final fractional atomic coordinates and isotopic thermal parameters, hydrogen atom positions, anisotropic thermal parameters, bond distances and bond angles are given below (**Tables 1 to 7**). The asymmetric unit is shown in **Figure 1**, along with the labelling scheme used.

1. (a) Sheldrick G.M., SHELX86, a computer program for crystal structure determination, University of Goetingen, 1986.

   (b) Sheldrick G.M., SHELX76, a computer program for crystal structure determination, University of Cambridge, 1976.

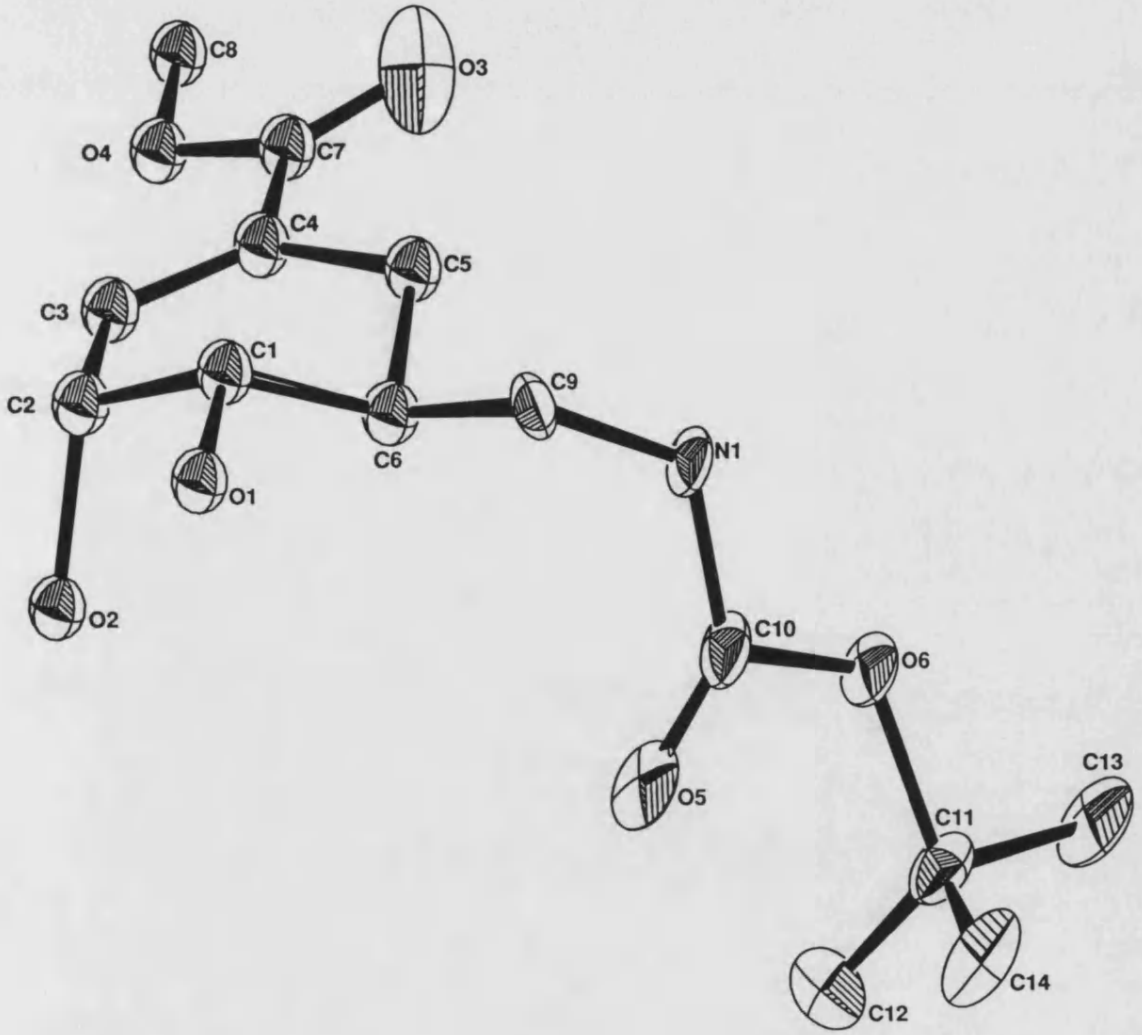


Figure 1

Table 1 Fractional atomic coordinates and thermal parameters (Å) for (179)

Atom	x	y	z	Uiso or Ueq	(***)
O3	0.1990(6)	-0.2697(5)	-0.3846(8)	0.090(6)	***
O5	0.3300(5)	-0.4468(4)	0.2100(8)	0.073(5)	***
O6	0.5130(5)	-0.3835(4)	0.1973(7)	0.051(4)	***
N1	0.3307(5)	-0.3041(4)	0.1182(8)	0.042(5)	***
C9	0.1899(6)	-0.2906(5)	0.0795(9)	0.040(5)	***
C10	0.3851(7)	-0.3839(6)	0.1772(11)	0.047(6)	***
C11	0.5978(8)	-0.4635(6)	0.2694(12)	0.054(7)	***
C12	0.5519(11)	-0.5551(7)	0.1990(14)	0.085(10)	***
C13	0.7273(8)	-0.4353(7)	0.2690(14)	0.081(9)	***
C14	0.5989(10)	-0.4659(9)	0.4042(14)	0.088(11)	***
O1	-0.0708(4)	-0.3577(3)	0.0222(6)	0.038(1)	
O2	-0.0878(4)	-0.4927(3)	-0.1652(6)	0.037(1)	
O4	0.0363(6)	-0.3594(4)	-0.5060(8)	0.066(2)	
C1	-0.0307(6)	-0.3331(5)	-0.0808(8)	0.031(2)	
C2	-0.1017(7)	-0.3944(5)	-0.2015(8)	0.032(2)	
C3	-0.0519(7)	-0.3789(5)	-0.3060(10)	0.037(2)	
C4	0.0613(6)	-0.3344(5)	-0.2896(8)	0.033(2)	
C5	0.1476(7)	-0.2999(5)	-0.1584(9)	0.036(2)	
C6	0.1158(6)	-0.3408(5)	-0.0480(8)	0.028(2)	
C7	0.1058(7)	-0.3173(6)	-0.3956(10)	0.044(2)	
C8	0.0749(10)	-0.3428(8)	-0.6120(12)	0.073(3)	

Table 2 Fractional atomic coordinates for the hydrogen atoms

Atom	x	y	z
H1	0.3758	-0.2622	0.0663
H11	-0.0567	-0.2595	-0.0994
H21	-0.2020	-0.3730	-0.2383
H31	-0.1013	-0.4005	-0.4056
H41	0.2462	-0.3185	-0.1442
H42	0.1389	-0.2239	-0.1563
H61	0.1452	-0.4141	-0.0358
H81	0.0119	-0.3803	-0.6958
H82	0.1731	-0.3671	-0.5882
H83	0.0699	-0.2678	-0.6324
H91	0.1598	-0.3193	0.1539
H92	0.1679	-0.2159	0.0686
H121	0.6135	-0.6119	0.2519
H122	0.5557	-0.5509	0.1038
H123	0.4536	-0.5685	0.1914
H131	0.7982	-0.4880	0.3191
H132	0.7546	-0.3679	0.3169
H133	0.7228	-0.4294	0.1709
H141	0.6595	-0.5231	0.4574
H142	0.5015	-0.4760	0.4014
H143	0.6363	-0.3995	0.4508

Table 3 Anisotropic thermal parameters (Å)

Atom	U11	U22	U33	U23	U13	U12
O3	0.075(5)	0.117(6)	0.078(8)	-0.009(5)	0.036(4)	-0.052(4)
O5	0.035(3)	0.057(4)	0.128(8)	0.046(4)	0.012(4)	-0.009(3)
O6	0.030(3)	0.046(3)	0.078(6)	0.018(3)	0.013(3)	-0.001(3)
N1	0.022(3)	0.042(4)	0.062(7)	0.013(4)	0.009(3)	-0.005(3)
C9	0.028(4)	0.032(4)	0.060(8)	-0.008(4)	0.015(4)	-0.005(3)
C10	0.029(5)	0.045(5)	0.068(9)	0.011(5)	0.002(4)	-0.009(4)
C11	0.044(5)	0.054(6)	0.064(10)	0.023(5)	0.008(5)	0.008(4)
C12	0.089(8)	0.059(7)	0.107(14)	-0.021(7)	0.016(7)	0.006(6)
C13	0.046(6)	0.080(7)	0.118(13)	0.043(7)	0.018(6)	0.013(5)
C14	0.068(7)	0.118(11)	0.078(14)	0.032(8)	0.002(7)	0.020(7)

Table 4 Bond lengths (Å)

O1	-C1	1.418(10)	O2	-C2	1.440(8)
O3	-C7	1.203(9)	O4	-C7	1.331(11)
O4	-C8	1.416(13)	O5	-C10	1.209(10)
O6	-C10	1.354(9)	O6	-C11	1.503(10)
N1	-C9	1.475(8)	N1	-C10	1.334(10)
C1	-C2	1.552(11)	C1	-C6	1.535(9)
C2	-C3	1.477(13)	C3	-C4	1.357(10)
C4	-C5	1.507(11)	C4	-C7	1.458(12)
C5	-C6	1.513(12)	C6	-C9	1.532(11)
C11	-C12	1.503(13)	C11	-C13	1.494(12)
C11	-C14	1.498(17)			

Table 5 Bond angles (°)

C8	-O4	-C7	116.6(8)	C11	-O6	-C10	120.0(7)
C10	-N1	-C9	119.6(7)	C2	-C1	-O1	110.8(6)
C6	-C1	-O1	114.1(6)	C6	-C1	-C2	109.0(6)
C1	-C2	-O2	109.0(6)	C3	-C2	-O2	109.4(6)
C3	-C2	-C1	112.6(6)	C4	-C3	-C2	123.9(8)
C5	-C4	-C3	120.7(8)	C7	-C4	-C3	122.6(8)
C7	-C4	-C5	116.7(7)	C6	-C5	-C4	114.3(6)
C5	-C6	-C1	108.7(6)	C9	-C6	-C1	110.6(6)
C9	-C6	-C5	111.8(6)	O4	-C7	-O3	122(1)
C4	-C7	-O3	122.9(9)	C4	-C7	-O4	115.0(8)
C6	-C9	-N1	111.3(7)	O6	-C10	-O5	124.7(8)
N1	-C10	-O5	125.7(7)	N1	-C10	-O6	109.6(7)
C12	-C11	-O6	110.0(8)	C13	-C11	-O6	102.3(7)
C13	-C11	-C12	112(1)	C14	-C11	-O6	109.1(9)
C14	-C11	-C12	112(1)	C14	-C11	-C13	111(1)

Table 6 Intermolecular distances (Å)

O1	...H81	2.95	1	0.0	0.0	-1.0
O1	...O2	2.84	-1	0.0	-1.0	0.0
O1	...N1	2.90	-2	1.0	0.0	0.0
O1	...H1	1.92	-2	1.0	0.0	0.0
O2	...O5	2.69	-1	0.0	-1.0	0.0
O2	...H61	2.86	-1	0.0	-1.0	0.0
O2	...H81	2.70	-1	0.0	-1.0	-1.0
O2	...H91	2.79	-1	0.0	-1.0	0.0

O3	...H121	2.66	-1	1.0	-1.0	0.0
O3	...H21	2.57	-2	0.0	0.0	0.0
O3	...H143	2.94	-2	1.0	0.0	1.0
O5	...H21	2.99	-1	0.0	-1.0	0.0
O6	...H83	2.78	-2	0.0	0.0	-1.0
O6	...H92	2.97	-2	0.0	0.0	0.0
H1	...C1	2.61	-2	0.0	0.0	0.0
C7	...H131	2.96	-1	1.0	-1.0	0.0
H92	...C13	2.99	-2	1.0	0.0	0.0
C14	...H142	2.89	-1	1.0	-1.0	1.0

Table 7 Intramolecular distances (Å)

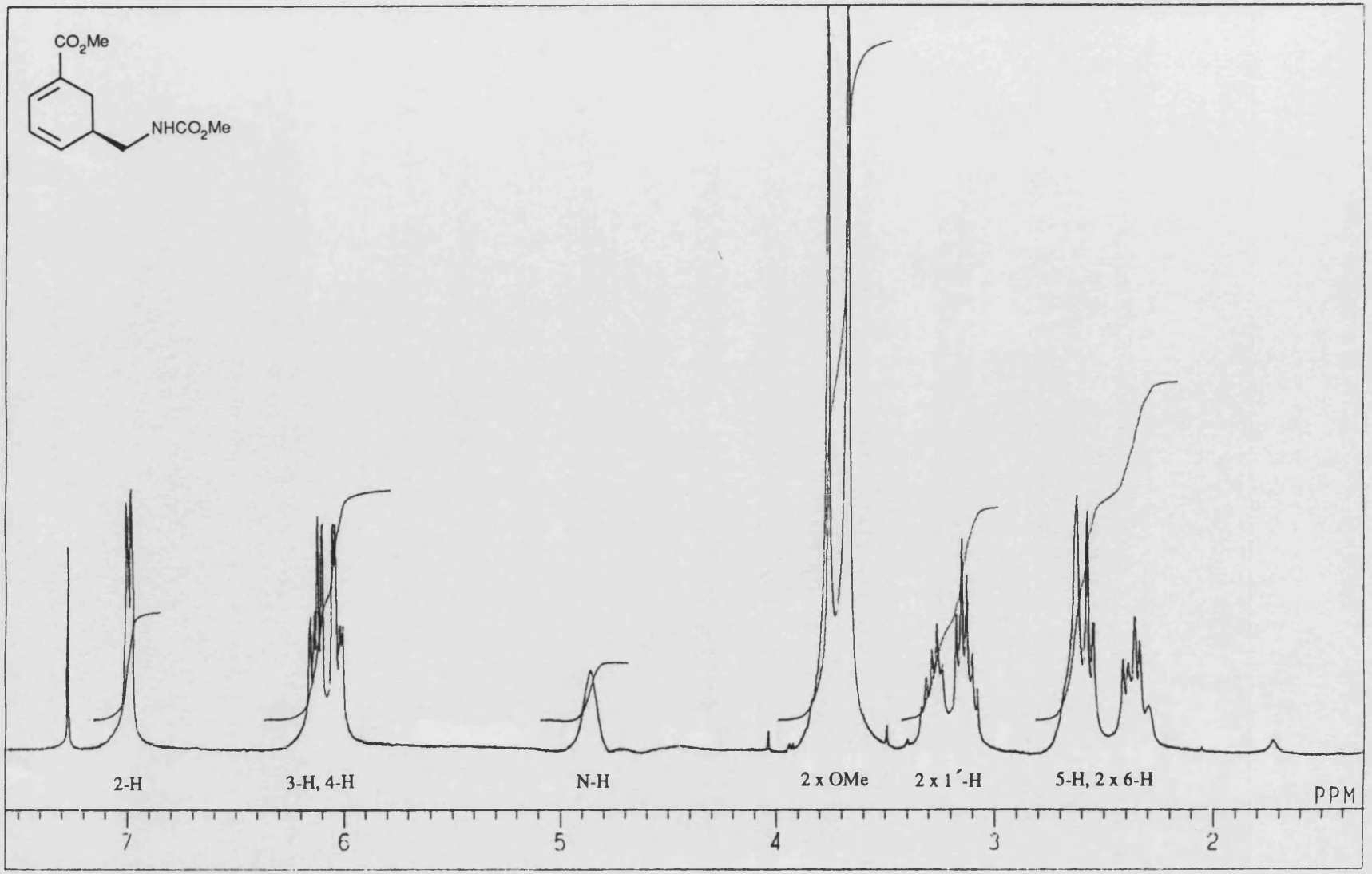
O1	...O2	2.78	O1	...H11	1.99
O1	...C2	2.45	O1	...H21	2.74
O1	...C6	2.48	O1	...H61	2.82
O1	...C9	2.89	O1	...H91	2.50
O2	...C1	2.44	O2	...H21	2.09
O2	...C3	2.38	O2	...H31	2.93
O2	...H61	2.70	O3	...O4	2.22
O3	...C4	2.34	O3	...C5	2.81
O3	...H41	2.63	O3	...H42	2.93
O3	...C8	2.62	O3	...H82	2.58
O3	...H83	2.61	O4	...C3	2.76
O4	...H31	2.27	O4	...C4	2.35
O4	...H81	2.05	O4	...H82	2.04
O4	...H83	2.05	O5	...O6	2.27
O5	...N1	2.26	O5	...H61	2.80

O5	...C9	2.79	O5	...H91	2.52
O5	...C11	2.81	O5	...C12	2.94
O5	...H123	2.26	O5	...C14	2.99
O5	...H142	2.32	O6	...N1	2.20
O6	...H1	2.40	O6	...C12	2.46
O6	...H122	2.70	O6	...H123	2.69
O6	...C13	2.33	O6	...H132	2.53
O6	...H133	2.54	O6	...C14	2.44
O6	...H142	2.67	O6	...H143	2.66
N1	...H41	2.73	N1	...C6	2.48
N1	...H61	2.65	N1	...H91	2.09
N1	...H92	2.10	H1	...C5	2.89
H1	...C6	2.92	H1	...C9	2.16
H1	...C10	2.10	C1	...H21	2.14
C1	...C3	2.52	C1	...C4	2.86
C1	...C5	2.48	C1	...H42	2.79
C1	...H61	2.16	C1	...C9	2.52
C1	...H91	2.70	C1	...H92	2.77
H11	...C2	2.18	H11	...C3	2.87
H11	...C5	2.64	H11	...C6	2.12
H11	...C9	2.77	C2	...H31	2.28
C2	...C4	2.50	C2	...C5	2.95
C2	...C6	2.51	C2	...H61	2.70
H21	...C3	2.07	C3	...C5	2.49
C3	...C6	2.85	C3	...C7	2.47
H31	...C4	2.02	H31	...C7	2.55
C4	...H41	2.11	C4	...H42	2.11
C4	...C6	2.54	C4	...H61	2.87

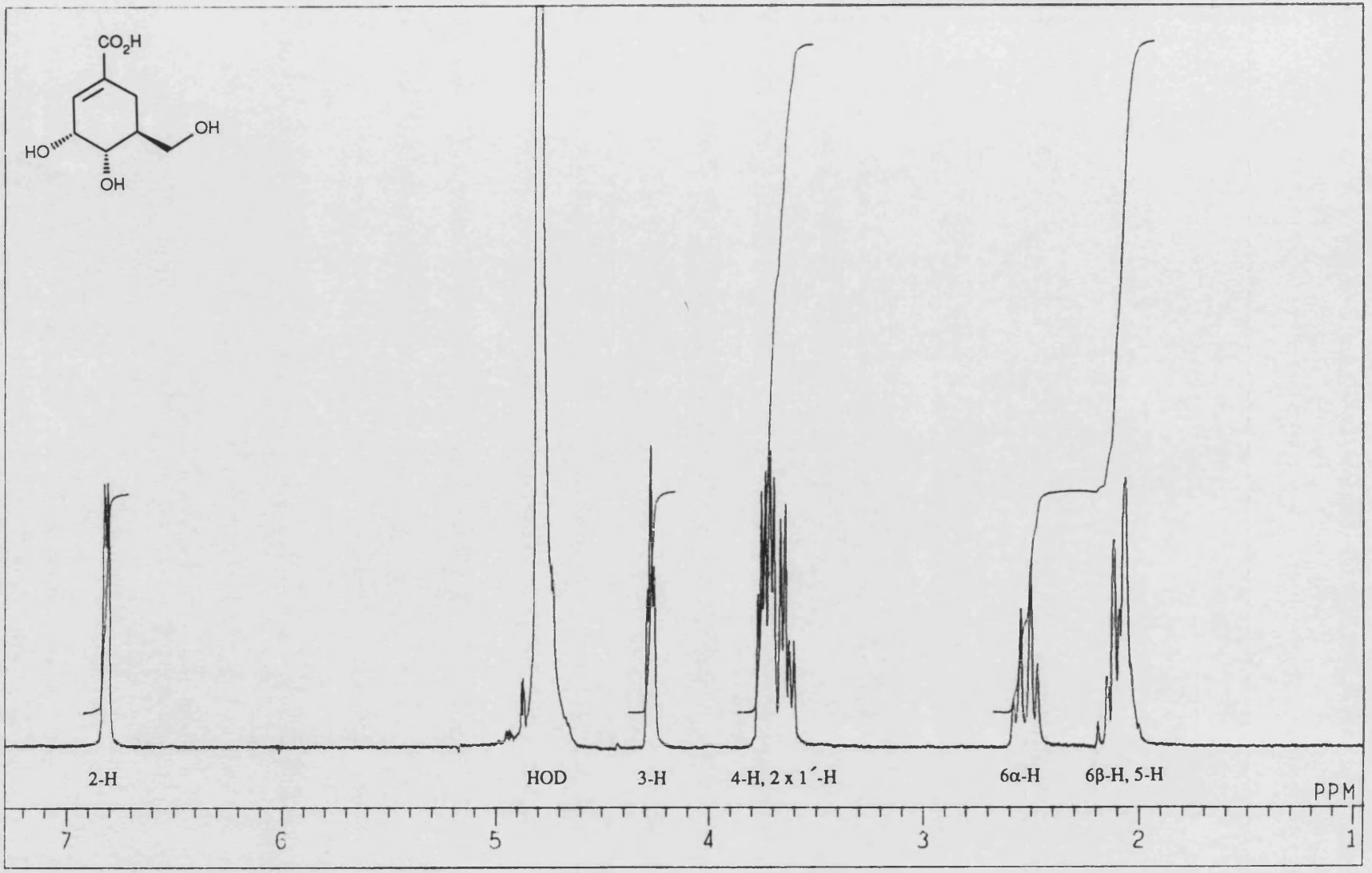
C5	...H61	2.12	C5	...C7	2.52
C5	...C9	2.52	C5	...H92	2.73
H41	...C6	2.12	H41	...C7	2.66
H41	...C9	2.81	H42	...C6	2.12
H42	...C7	2.88	H42	...C9	2.65
C6	...H91	2.14	C6	...H92	2.14
H61	...C9	2.12	H61	...C10	2.88
C7	...C8	2.34	C7	...H82	2.61
C7	...H83	2.62	C9	...C10	2.43
H91	...C10	2.59	C10	...C11	2.48
C10	...H123	2.70	C10	...C14	3.00
C10	...H142	2.69	C11	...H121	2.12
C11	...H122	2.13	C11	...H123	2.12
C11	...H131	2.12	C11	...H132	2.11
C11	...H133	2.12	C11	...H141	2.13
C11	...H142	2.12	C11	...H143	2.11
C12	...C13	2.48	C12	...H131	2.74
C12	...H133	2.70	C12	...C14	2.49
C12	...H141	2.72	C12	...H142	2.75
H121	...C13	2.77	H121	...C14	2.72
H122	...C13	2.67	H123	...C14	2.75
C13	...C14	2.46	C13	...H141	2.77
C13	...H143	2.62	H131	...C14	2.72
H132	...C14	2.66			

APPENDIX THREE

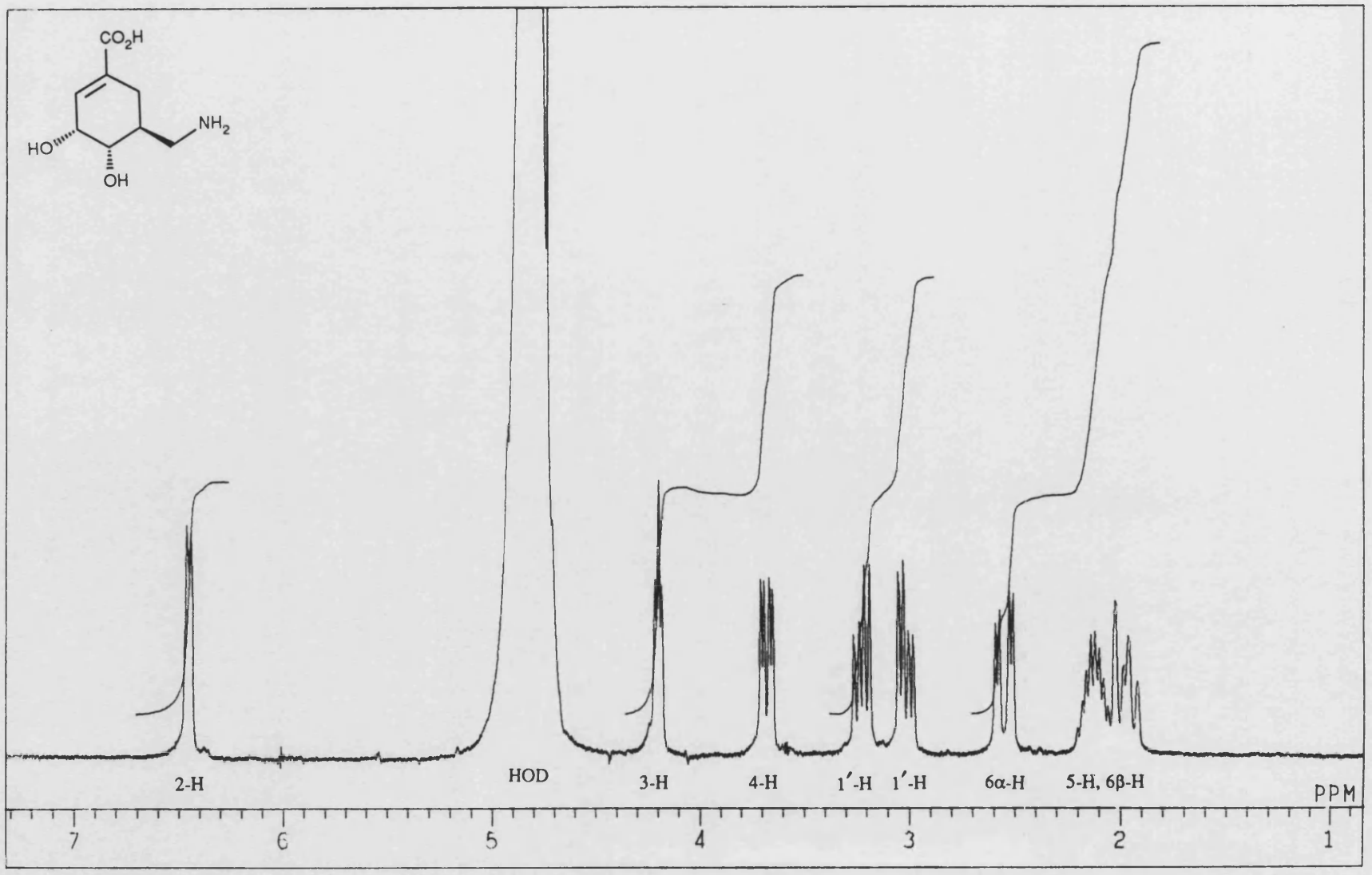
SELECTED NMR SPECTRA



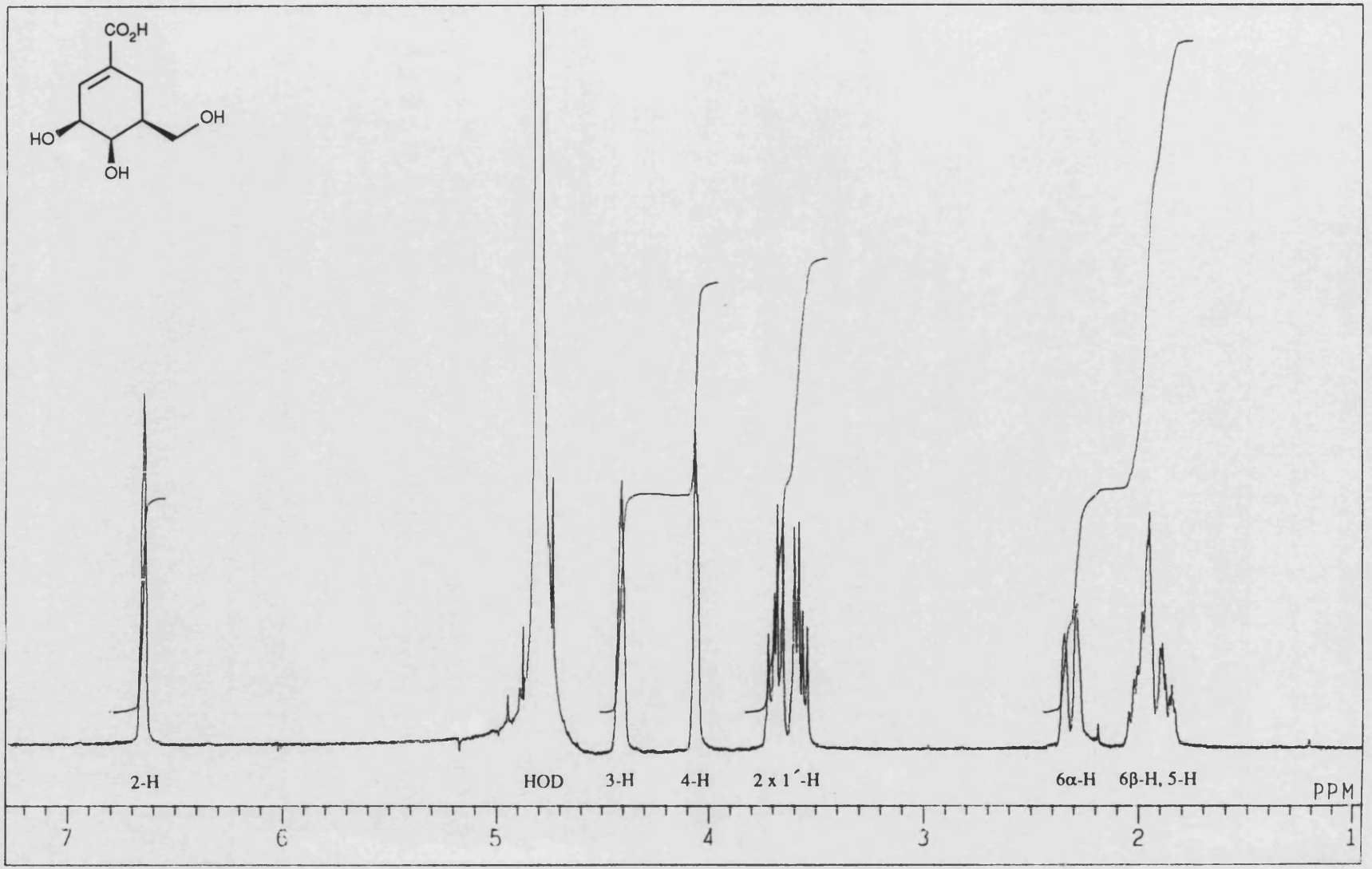
^{1}H NMR Spectrum of (127) (270 MHz, $CDCl_3$)



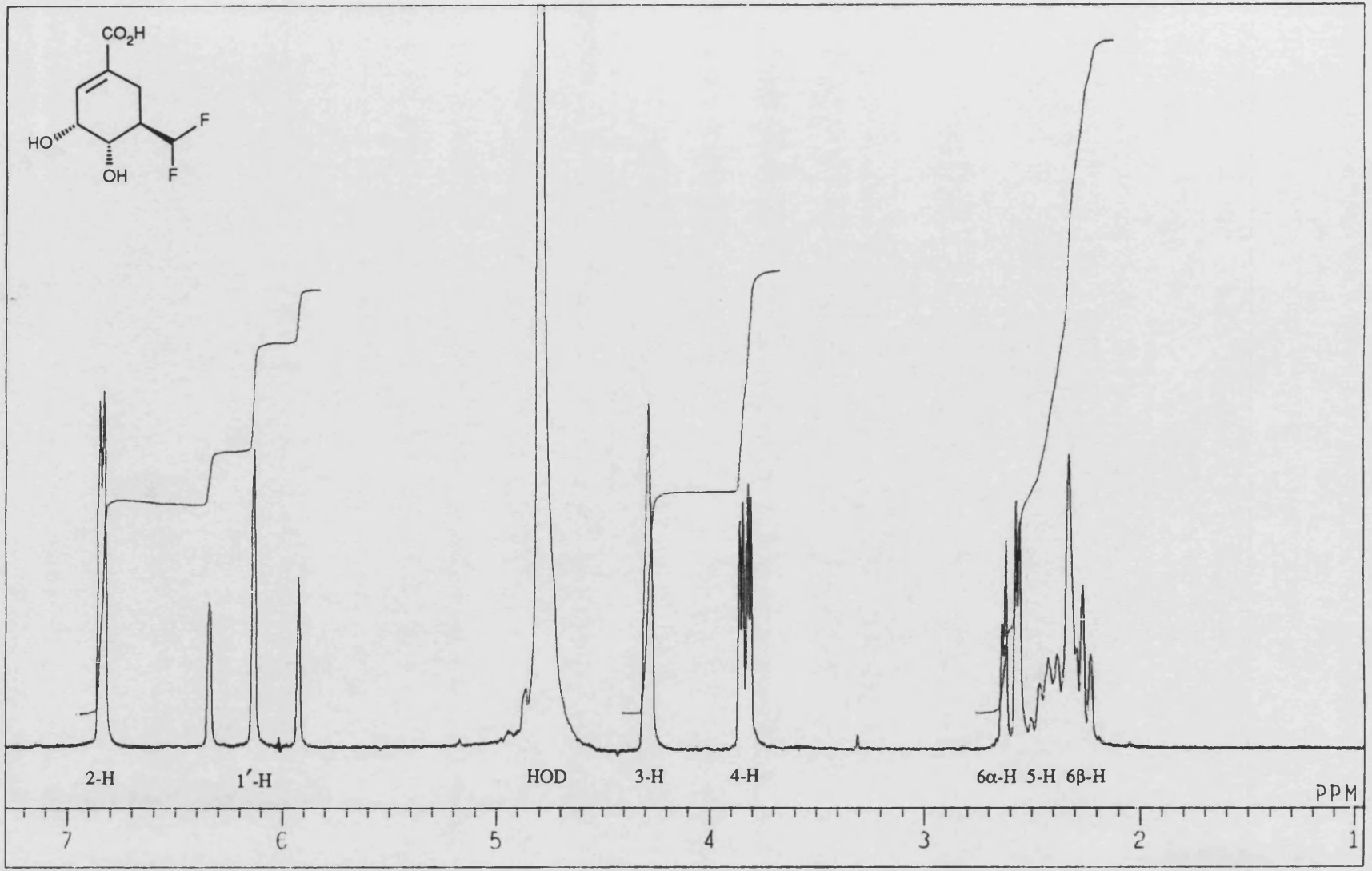
^{1}H NMR Spectrum of (174) (270 MHz, D_2O)



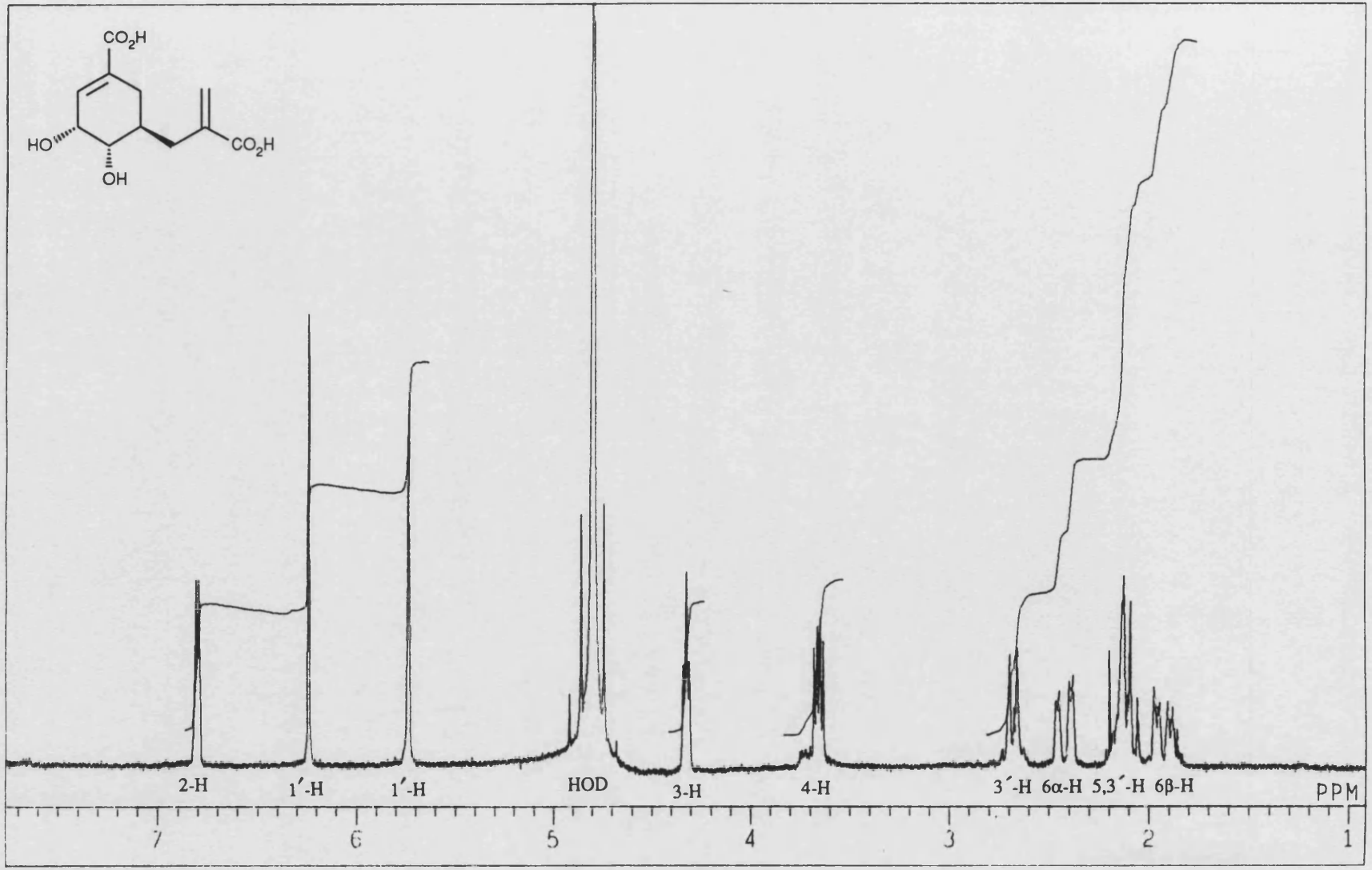
^{1}H NMR Spectrum of (181) (270 MHz, D_2O)



^{1}H NMR Spectrum of (196) (270 MHz, D_2O)



^{1}H NMR Spectrum of (203) (270 MHz, D_2O)



^{1}H NMR Spectrum of (212) (270 MHz, D_2O)

APPENDIX FOUR

PUBLICATION

Malcolm M. Campbell, Mary F. Mahon, Malcolm Sainsbury, Philip A. Searle and Gareth M. Davies,

Synthesis of (±)-Homogabaculine and (±)-Homoshikimic Acid,

Tetrahedron Lett., 1991, **32**, 951.

Tetrahedron Letters, Vol.32, No.7, pp 951-954, 1991
Printed in Great Britain

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SYNTHESIS OF (±)-HOMOGABACULINE AND (±)-HOMOSHIKIMIC ACID

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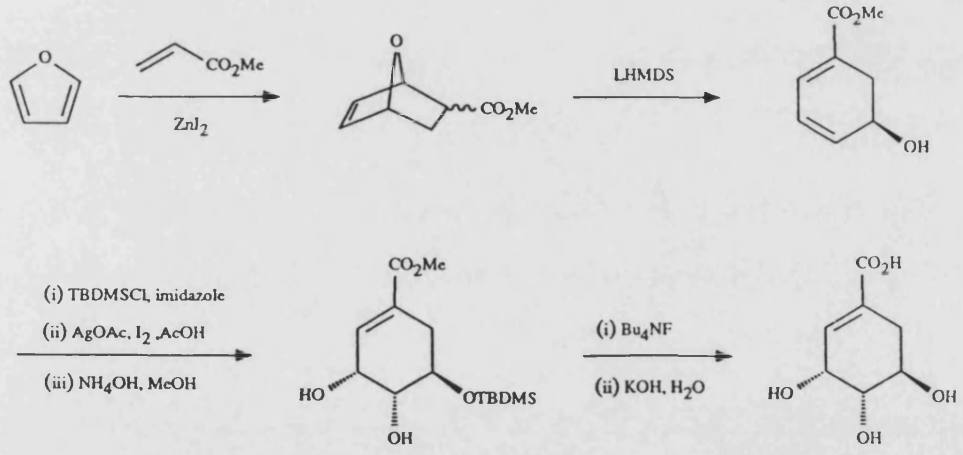
Summary: Syntheses of homogabaculine and homoshikimic acid are described utilising the base mediated ring opening of the cycloadducts of methyl acrylate and *N*-carbamoyl-1,2-dihydropyridines.

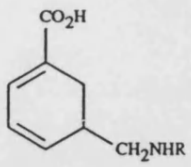
We have developed a brief and stereocontrolled synthesis of shikimic acid (1) involving a Diels Alder reaction between furan and methyl acrylate, followed by ring opening of the adducts with lithium hexamethyldisilazide (LHMDS) (scheme 1)[1] (for chiral routes based upon this strategy and intermediates see references 2a and 2b).



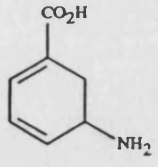
(1)

Scheme 1 Synthesis of shikimic acid

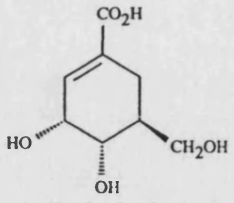
We now describe a further cycloaddition sequence leading to the homologue (2, R=H) of gabaculine (3) and the homologue (4) of shikimic acid (1) as potential enzyme substrates.



(2)

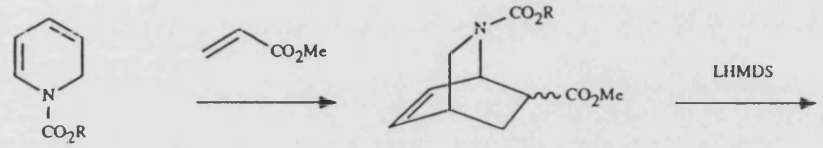


(3)

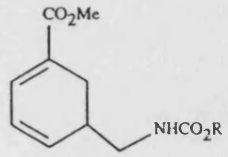


(4)

It is known[3] that the 1,2-dihydropyridine (5, R=Me) reacts with methyl acrylate to form the mixed *exo* and *endo* adducts (6, R=Me), in the ratio 7:5. The ^{t}butyl adducts (6, R=^{t}Bu) are similarly produced from the 1,2-dihydropyridine (5, R=^{t}Bu)[4], but now in the ratio 1:1. When treated with LHMDS at -78°C in THF the adducts (6, R=Me) afford the methyl 5,6-dihydrobenzoate (7), which can be hydrolysed with 2M sodium hydroxide in refluxing THF/H_2O to give homogabaculine (2, R=H) in 37% yield together with its carbamoyl derivative (2, R=CO_2Me), in 56% yield. Extended reaction periods tend to diminish the yield of products, while hydrolysis of the ester at 20°C gives only the carbamoyl compound (2, R=CO_2Me), in 65% yield.



(5) (6)



(7)

NaOH, THF, H_2O → (2)

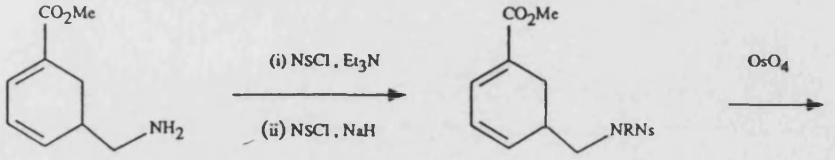
Methyl homogabaculine is formed directly from the adduct (6, R=^{t}Bu) by treatment with trifluoroacetic acid in 90% yield, and this compound when reacted with 4-nitrobenzenesulphonyl chloride (NsCl) in the presence of triethylamine affords the sulphonamide (8). Further treatment of this product with sodium hydride and 4-nitrobenzenesulphonyl chloride forms the disulphonimide (9)[5]. Overall yield for the two steps is 42%.

Osmylation[6] of the disulphonimide gives a mixture of the three diols (10), (11), and (12) in 16, 8,and 10% yields respectively, whereas a Prévost hydroxyacetoxylation reaction[1] affords two hydroxyacetates as an inseparable mixture, which on hydrolysis with aqueous ammonium hydroxide/methanol produces the diol

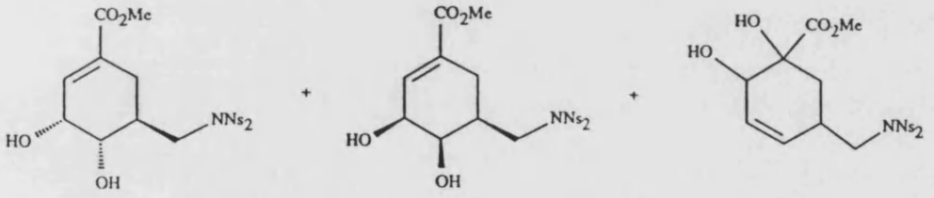
(10)[7] in 35% overall yield. This compound may then be protected, prior to treatment with potassium iodide in DMF at 130°C. This reaction leads to a mixture of the iodide (13), the diene (14), and the formate ester (15) in 23%, 9%, and 33% yields respectively, whereas treatment of the acetonide with potassium iodide in refluxing toluene containing 18-crown-6 gives only the iodide in 63% yield.

Hydrolysis of the formate ester to homoshikimic acid is accomplished in two steps: first, treatment with Amberlyst-15 acid ion exchange resin in methanol, followed by glacial acetic acid in aqueous THF at 60°C affords the triol (16) in 63% yield. This is reacted with 2M NaOH to give homoshikimic acid (4) in quantitative yield.

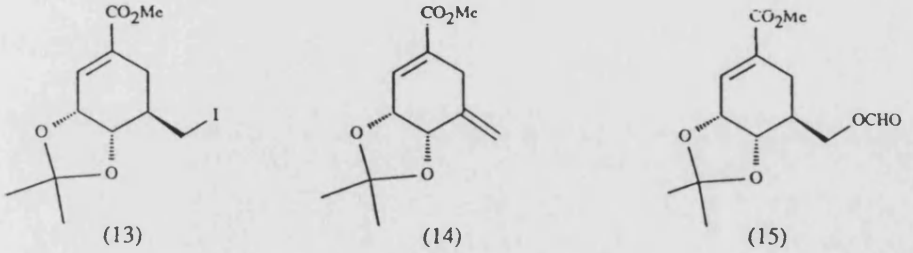
Alternatively the triol (16) can be prepared by treatment of the iodide (13) with sodium acetate in DMF at 110°C to give the acetate (17) (63%), hydrolysis to the alcohol (18) (aqueous ammonia, methanol, 81%) and removal of the acetonide group (acetic acid, aqueous THF, 55°C, 70%).



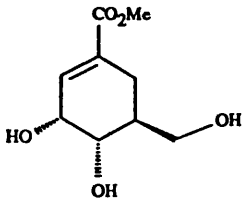
(8) R = H
(9) R = Ns



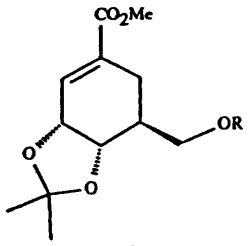
(10) (11) (12)



(13) (14) (15)



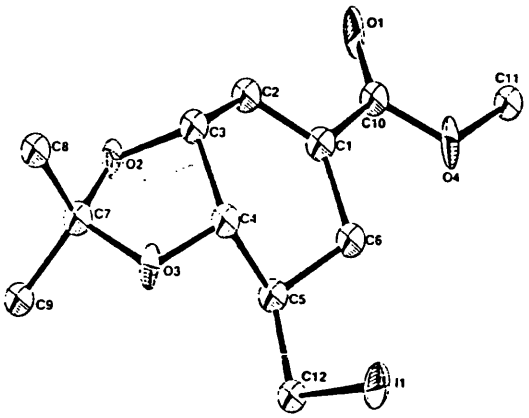
(16)



(17) R = Ac

(18) R = H

All the numbered compounds described are fully characterised. A single X-ray crystallographic determination of the iodide (13) has been carried out. Full data have been deposited at the Cambridge data bank. Relevant details are as follows:
the compound crystallised (from light petroleum ether) in space group P1 with a=5.660(2), b=8.252(4), c=15.014(3)A°, α=97.32(3), β=95.59(2), γ=94.34(3)°, U=689A°^{3}, and D_c=1.687gcm^3 for Z=2 at room temperature. The structure was solved by direct methods using 1704 unique reflections with I ≥ 3σI, and refined by full matrix least squares to final residuals of R=R_w=7.87% for unit weights.



ORTEP diagram of (13).

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References:

1. M.M.Campbell, M.Sainsbury, and R.Yavaradeh, *Tetrahedron*, **40**, 5063(1984); see also D.Rajapaksa, D.A.Keay, and H.Rodrigo, *Can.J.Chem.*, **62**, 826(1984).

2a. A.J.Birch, L.F.Kelley, and D.Weerasuria, *J.Org.Chem.*, **53**, 278 (1988); 2b. T.Takahashi, A.Iyobe, Y.Arai, and T.Koizumi, *Synthesis*, 189 (1989).

3. H. Sliwa, and I. Y. Lebot, *Tetrahedron Letters*, **18**, 4129 (1977).

4. R.J.Sundberg and J.D.Bloom, *J.Org.Chem.*, **46**, 4836 (1980).

5. P.J.DeChristopher, J.P.Adamek, G.D.Lyon, S.A.Klein, and R.J.Baumgarten *J.Org.Chem.*, **39**, 3525 (1974).

6. V.VanReenen,R.C.Kelly,and D.Y.Cha, *Tetrahedron Letters*, **17**, 1973 (1976).

7. Note that the stereochemical outcome of this Prévost reaction is not that expected from a 3,4-α-epiiodonium intermediate generated through steric approach control. A possible alternative mechanism which accounts for the observed stereochemistry of the product has been discussed previously: see reference 1.

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